Cotransfection of 293Cre cells with pBHG10lox and a "Lox" shuttle plasmid for generation of Ad expression vectors

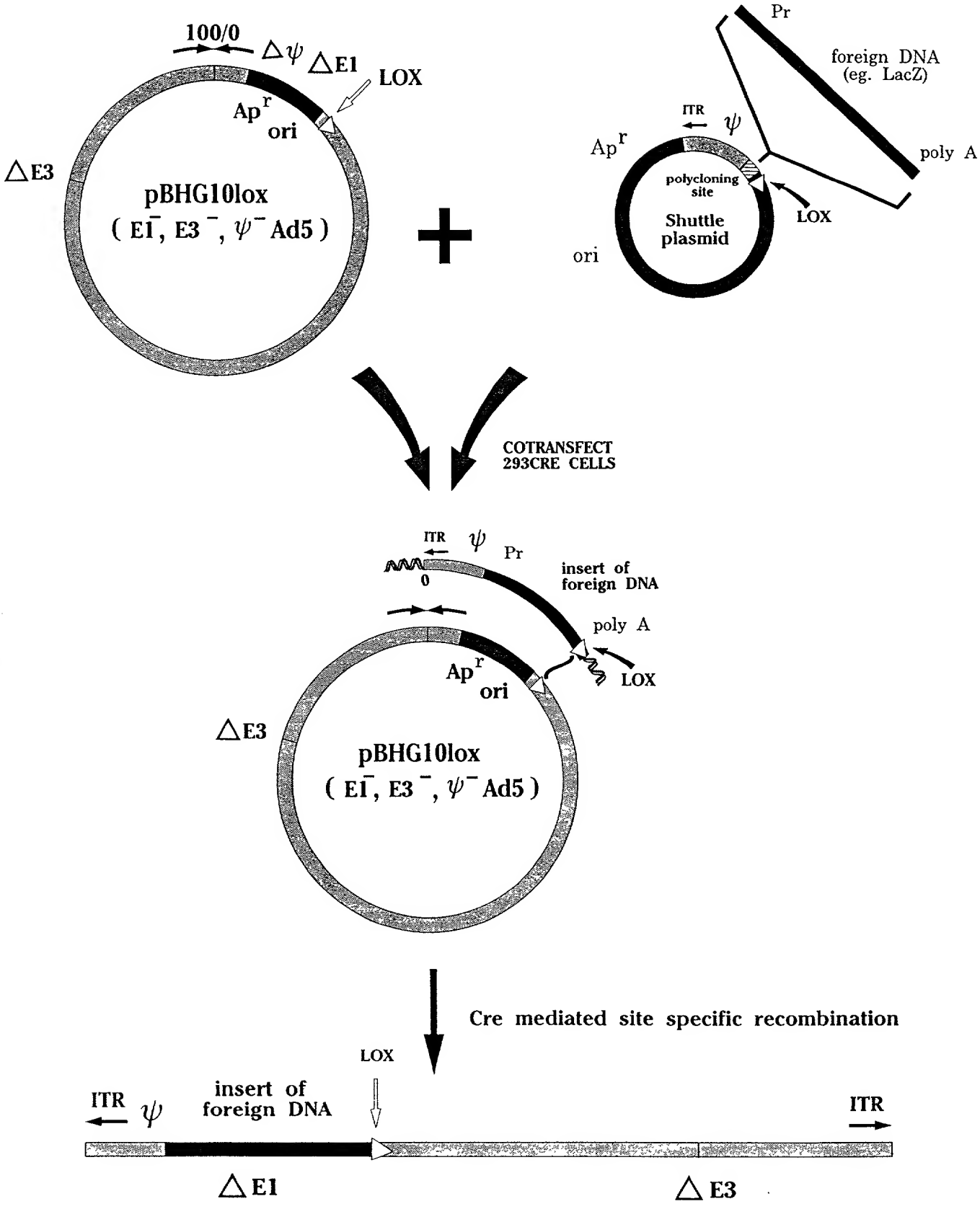
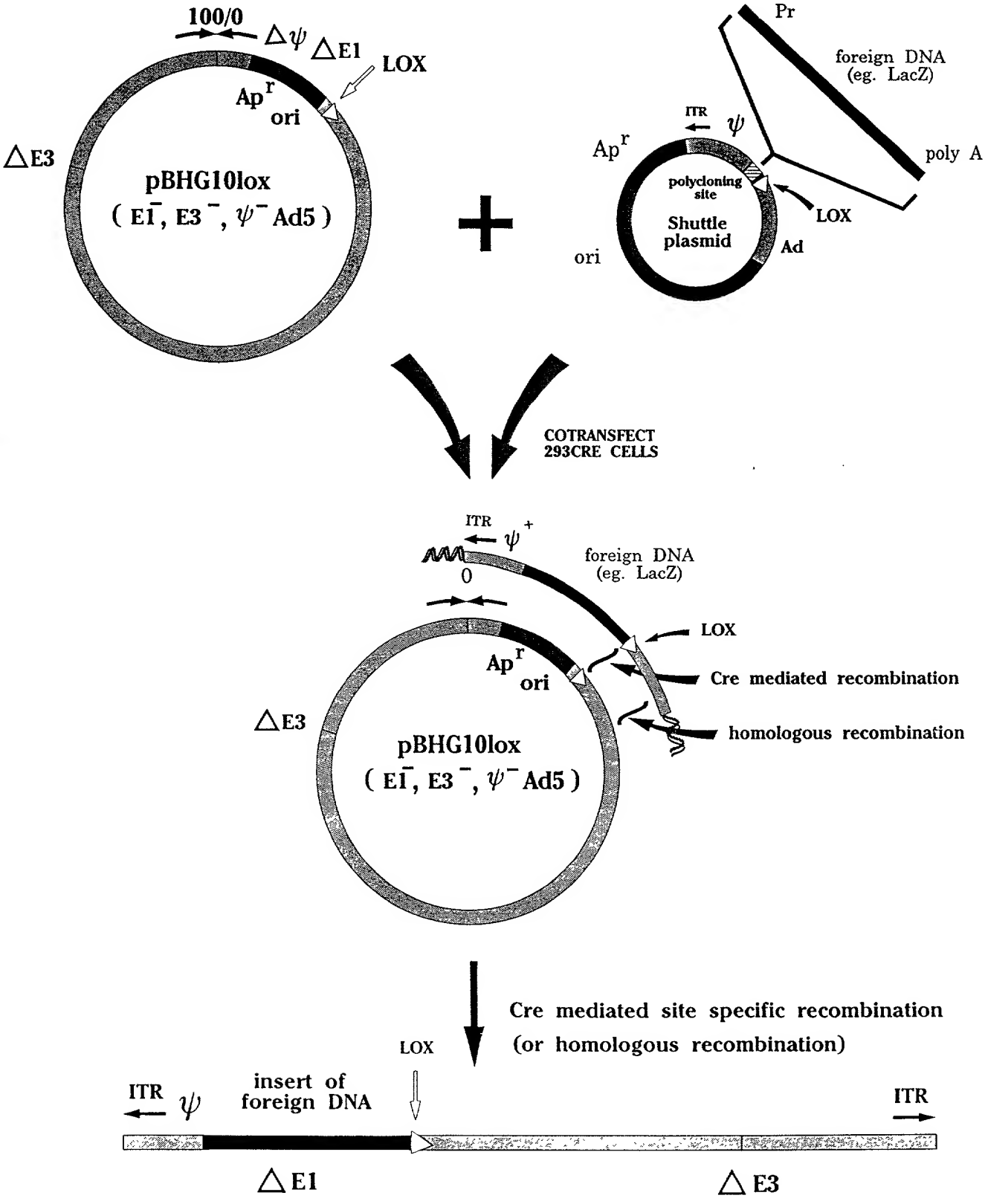


Fig. 1

Cotransfection of 293Cre cells with pBHG10lox and a "lox" shuttle plasmid for generation of Ad expression vectors



DEFECTIVE VIRAL VECTOR

Fig. 2A

CONSTRUCTION OF VARIOUS SHUTTLE PLASMIDS

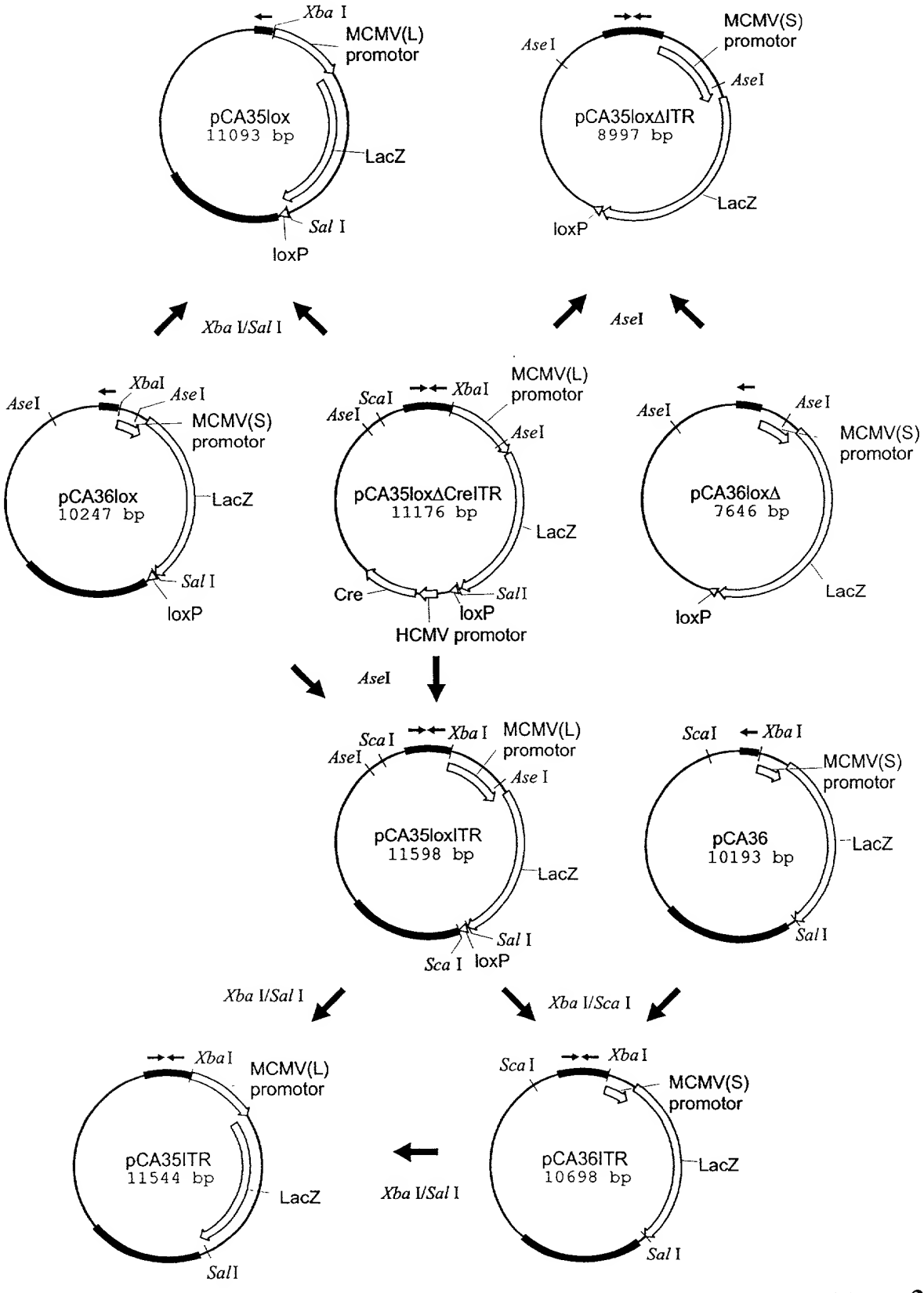


Figure 2B

OLIGONUCLEOTIDES USED IN CLONING

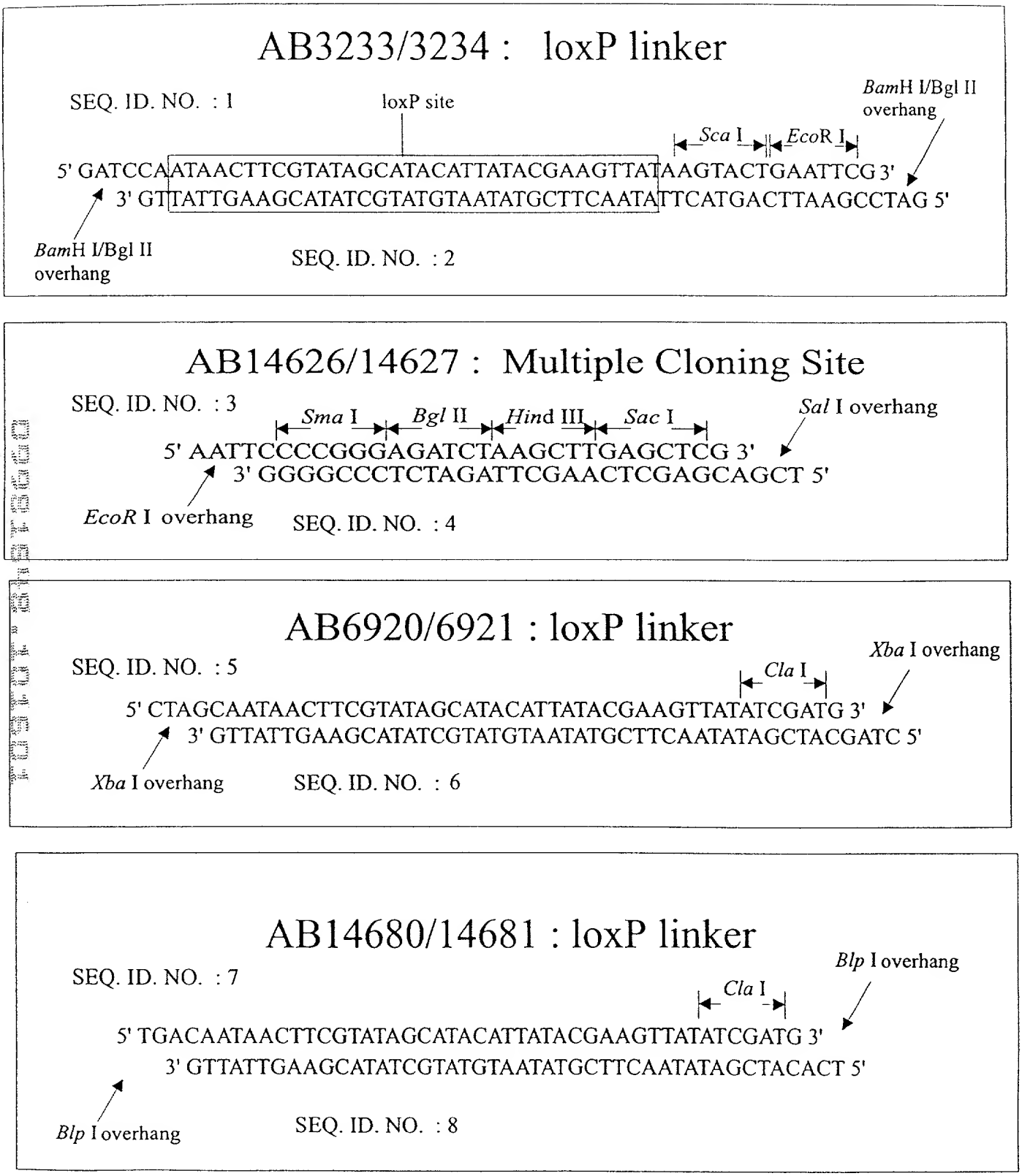


Fig. 3

CONSTRUCTION OF A CIRCULAR GENOMIC PLASMID FOR Ad VECTOR RESCUE USING THE Cre/ loxP SYSTEM

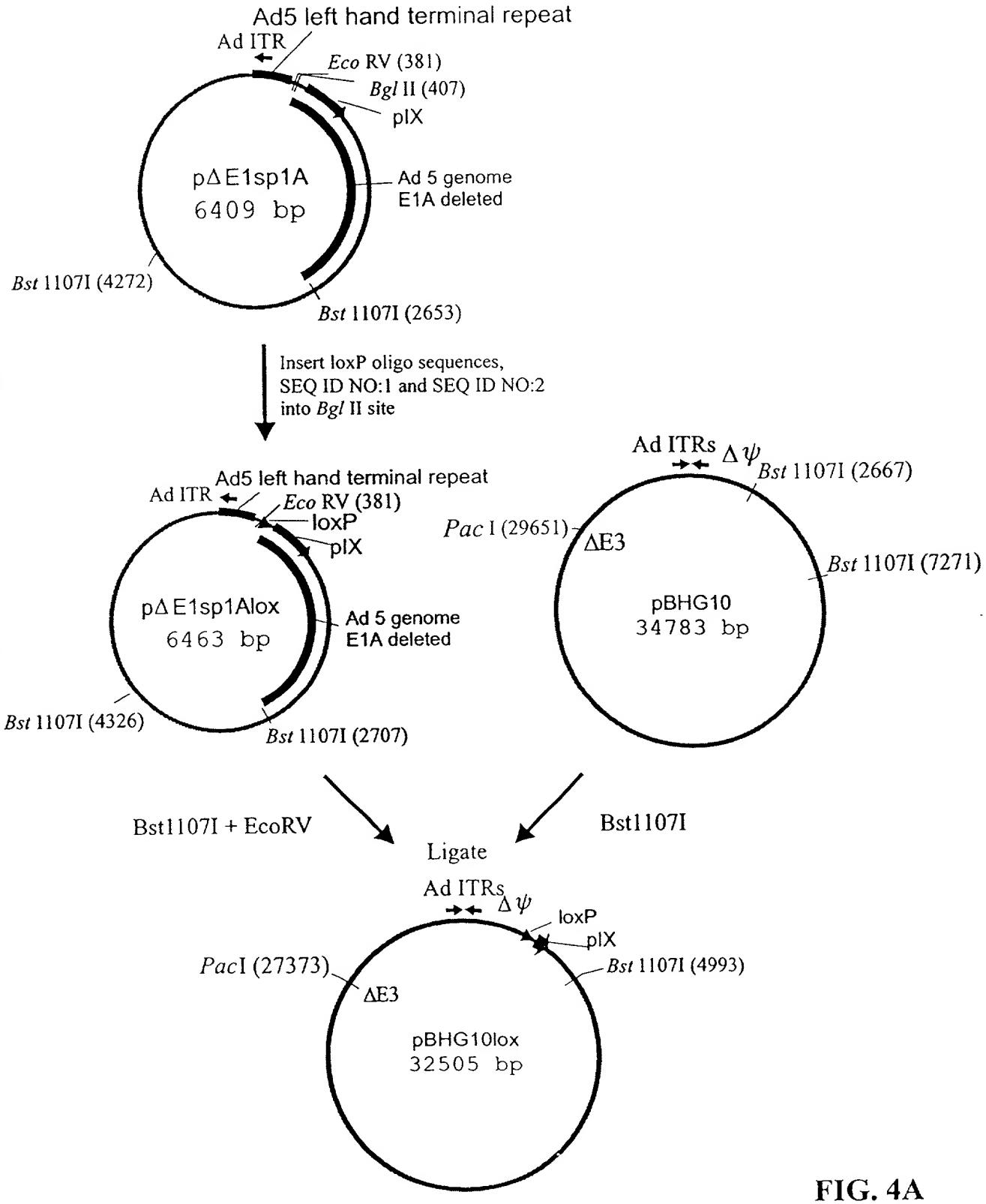


FIG. 4A

CONSTRUCTION OF pBHGdX1Plox

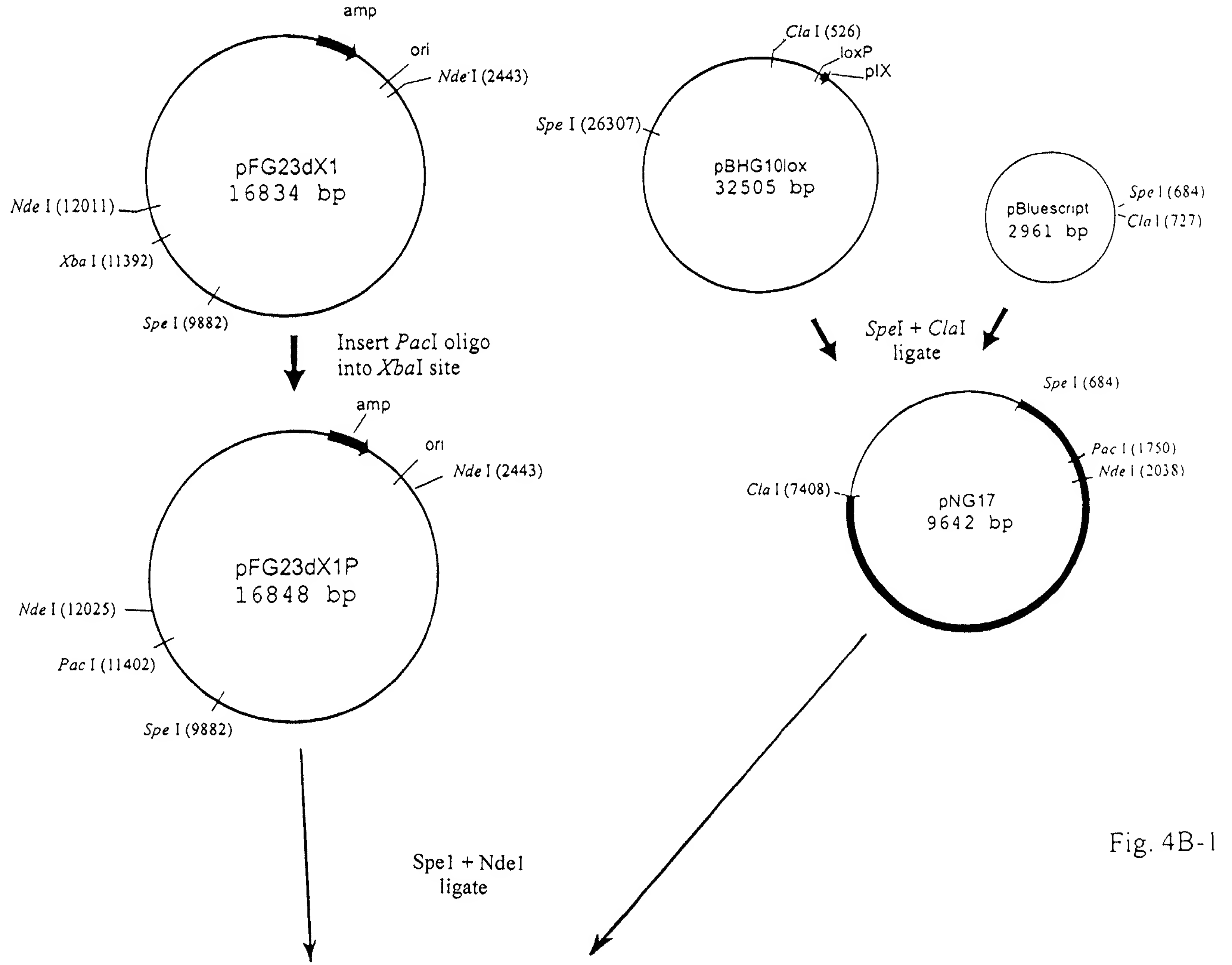


Fig. 4B-1

CONSTRUCTION OF pBHGdX1Plox

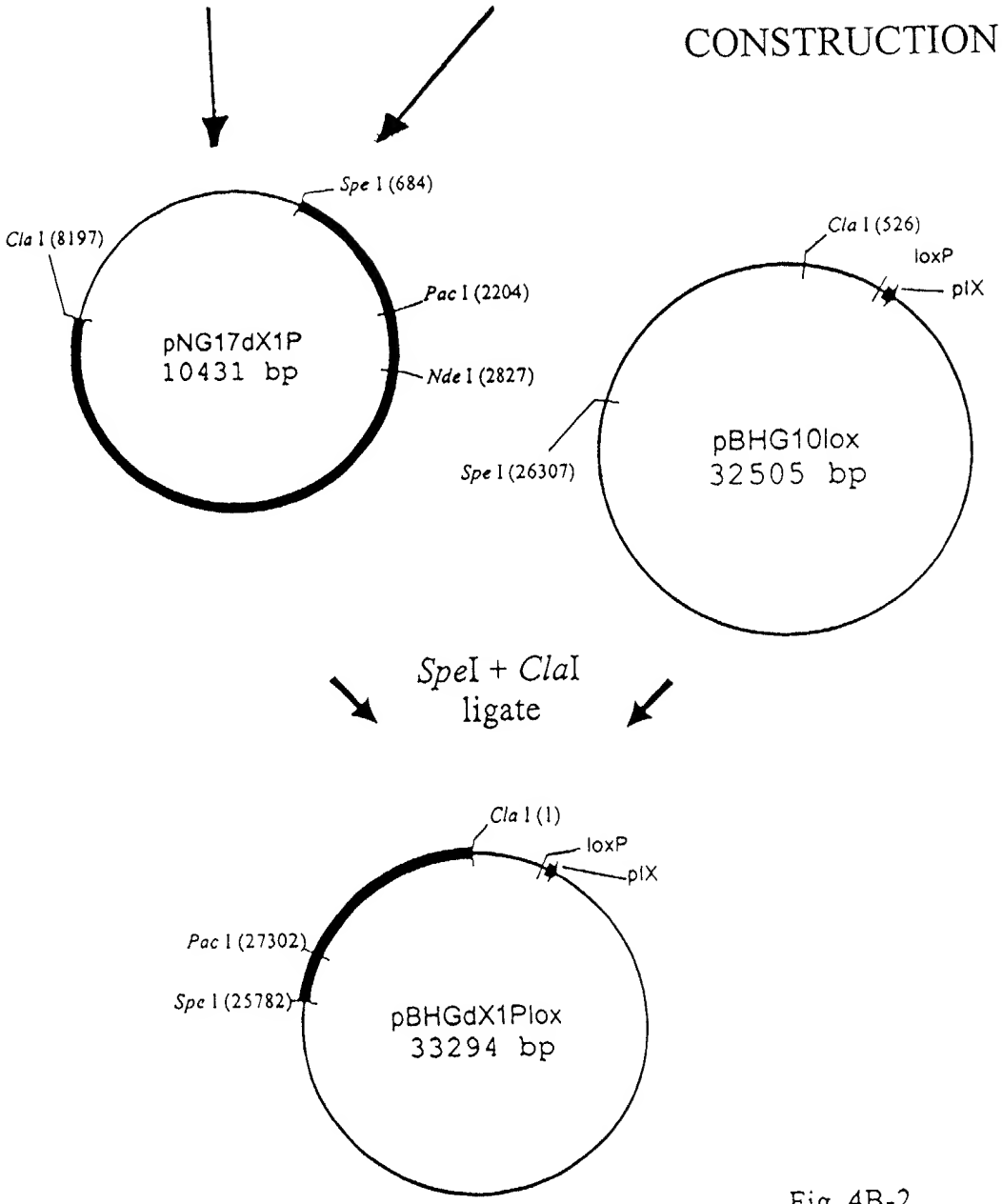


Fig. 4B-2

CONSTRUCTION OF pBHGE3lox

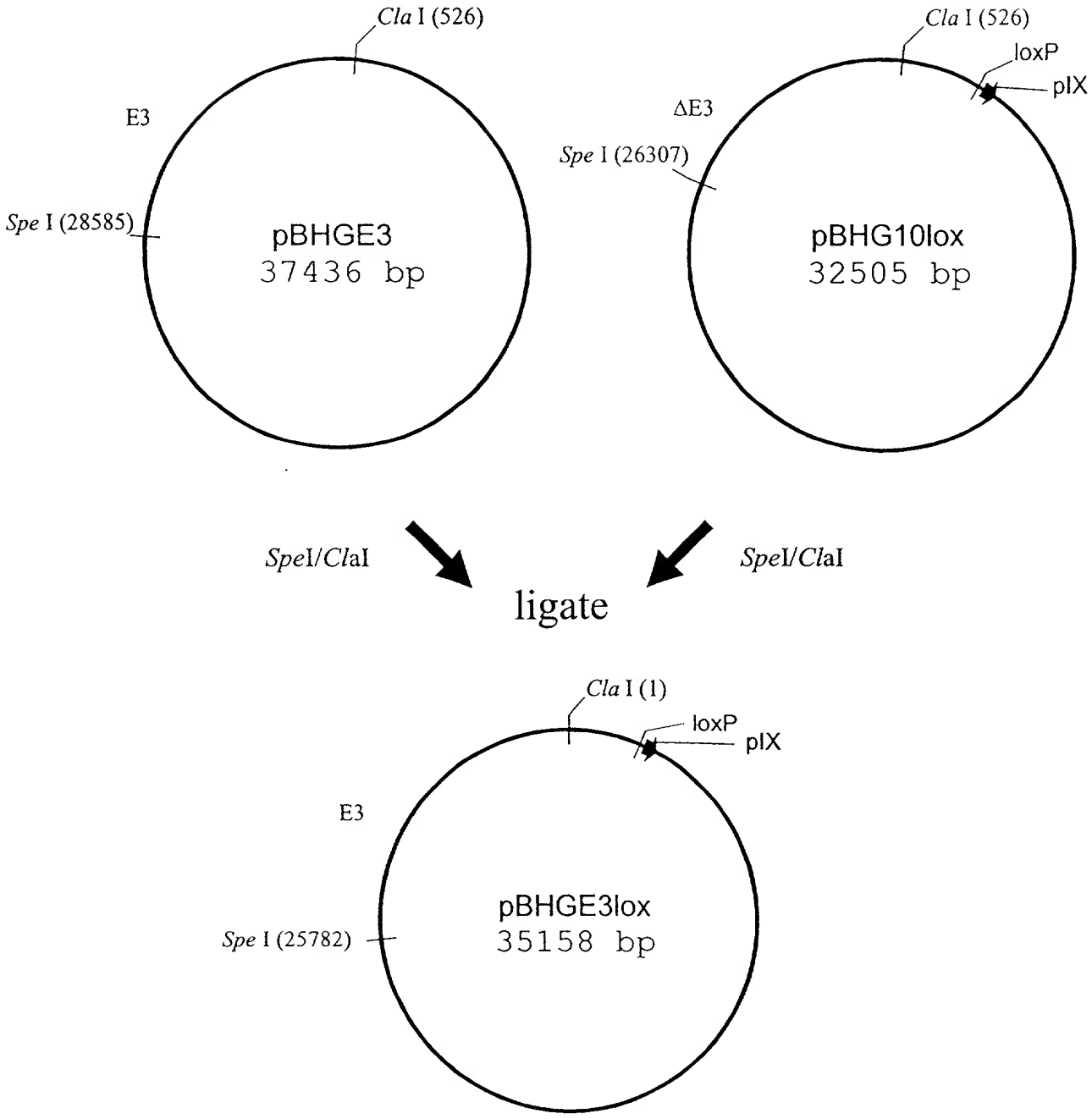


Fig. 4C

CONSTRUCTION OF Ad GENOMIC PLASMIDS ENCODING CRE

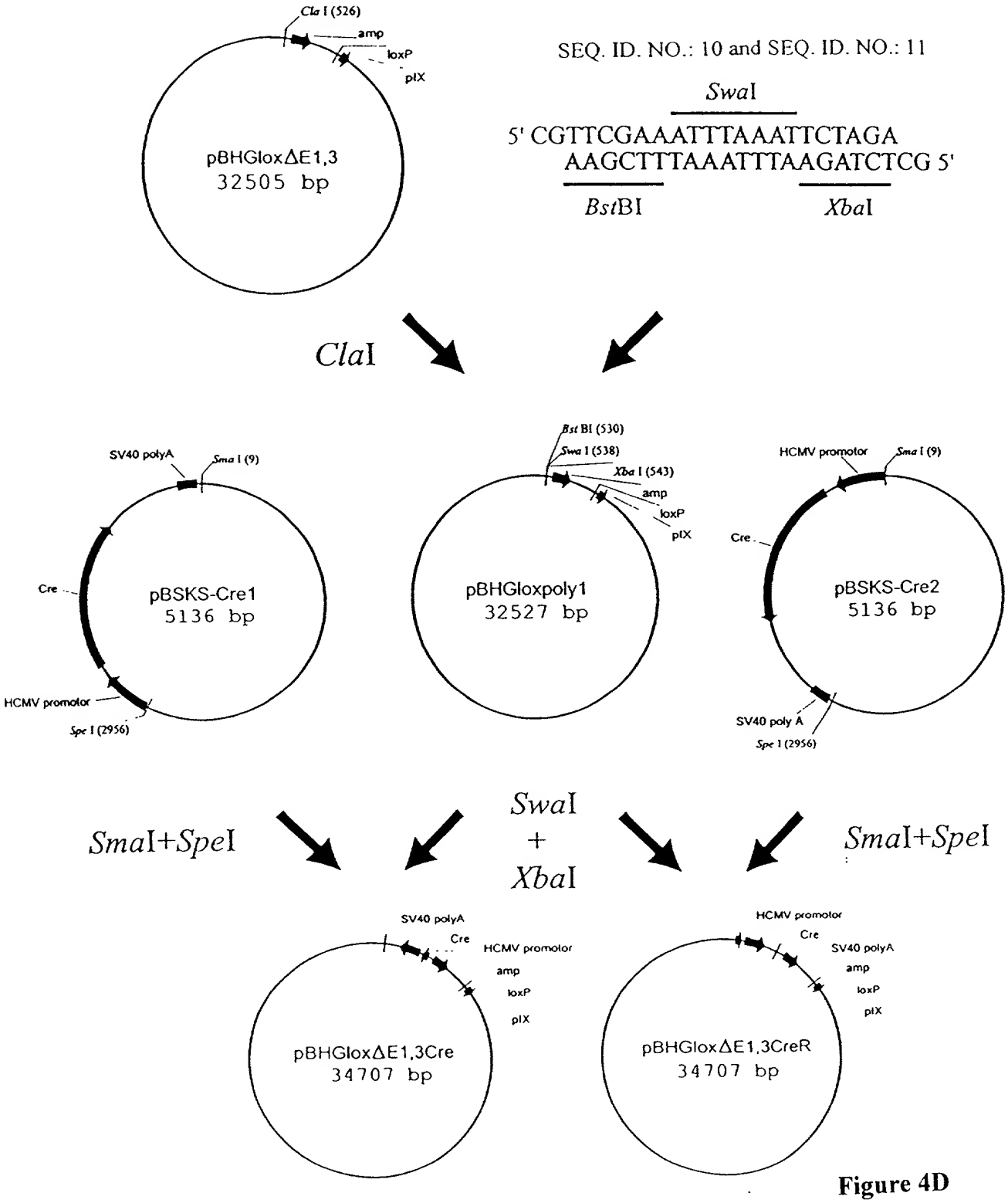


Figure 4D

CONSTRUCTION OF pΔE1SP1A & pΔE1SP1B loxP PLASMIDS FOR RESCUE OF FOREIGN DNA

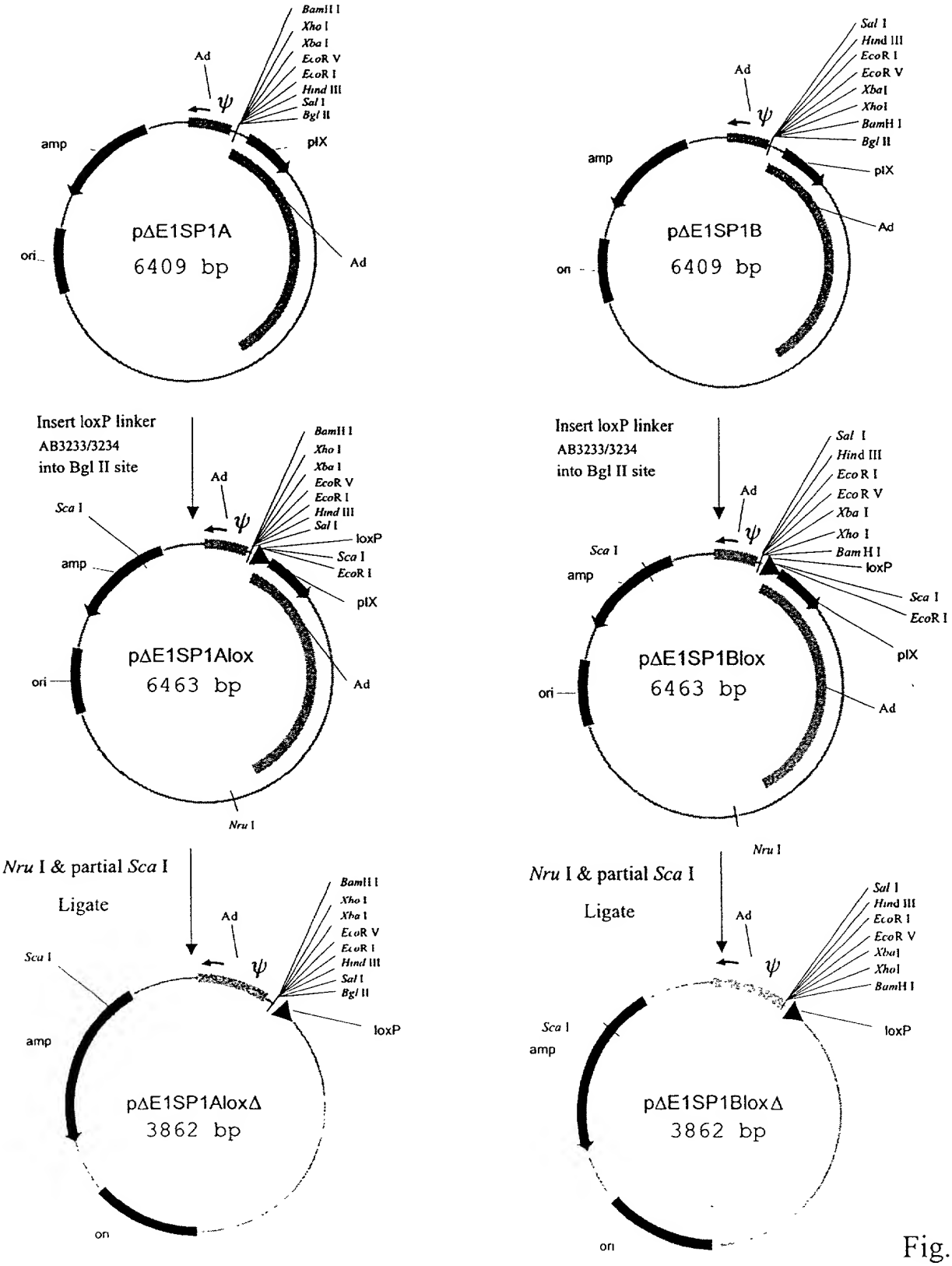


Fig. 5A

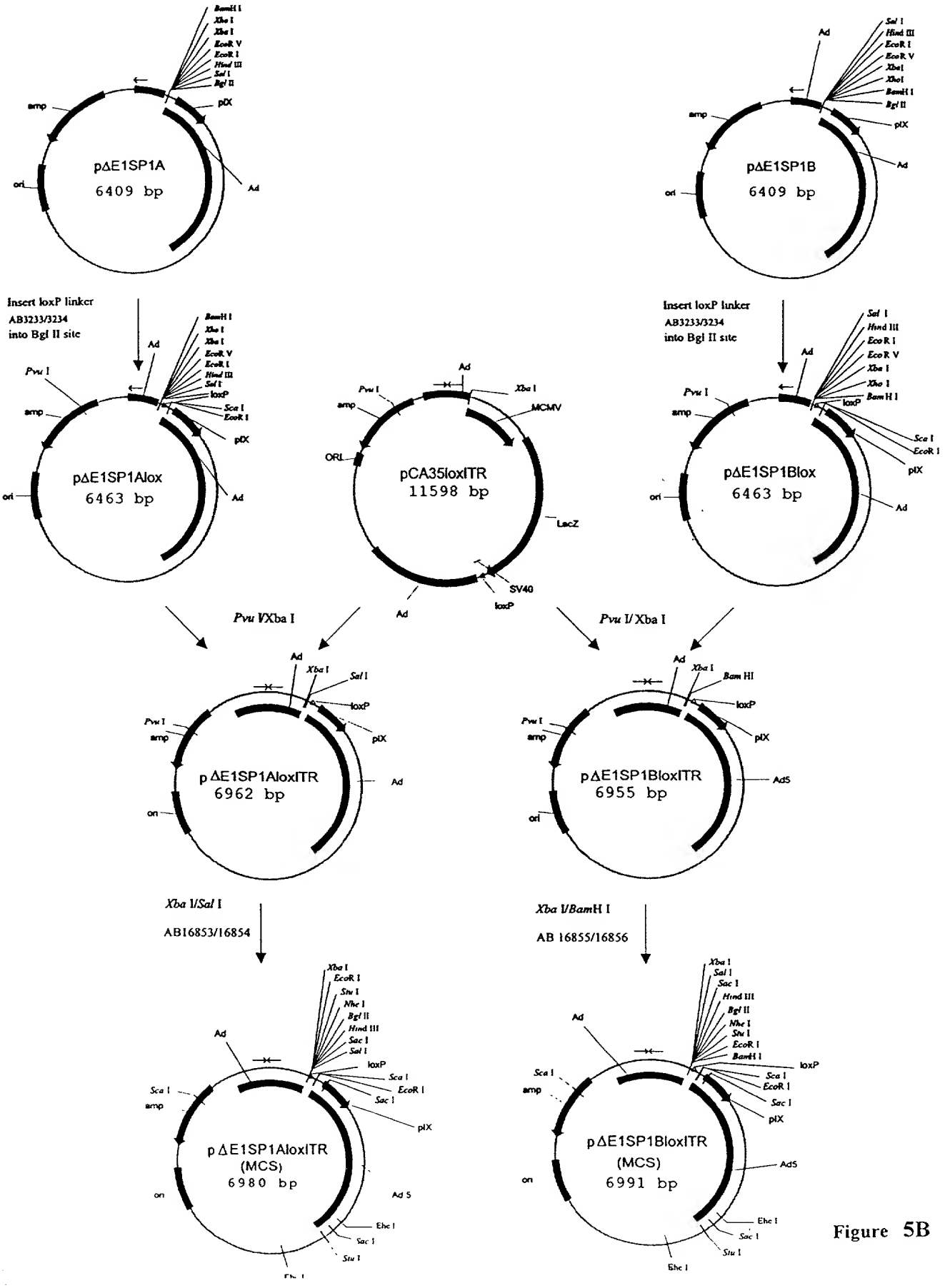

pΔE1SP1A
6409 bp
pΔE1SP1B
6409 bp
Insert loxP linker
AB3233/3234
into Bgl II site
pΔE1SP1Alox
6463 bp
pCA35loxITR
11598 bp
pΔE1SP1Blox
6463 bp
Pvu I/Xba I
pΔE1SP1AloxITR
6962 bp
pΔE1SP1BloxITR
6955 bp
Xba I/Sal I
AB16853/16854
Xba I/BamH I
AB 16855/16856
pΔE1SP1AloxITR
(MCS)
6980 bp
pΔE1SP1BloxITR
(MCS)
6991 bp

Figure 5B

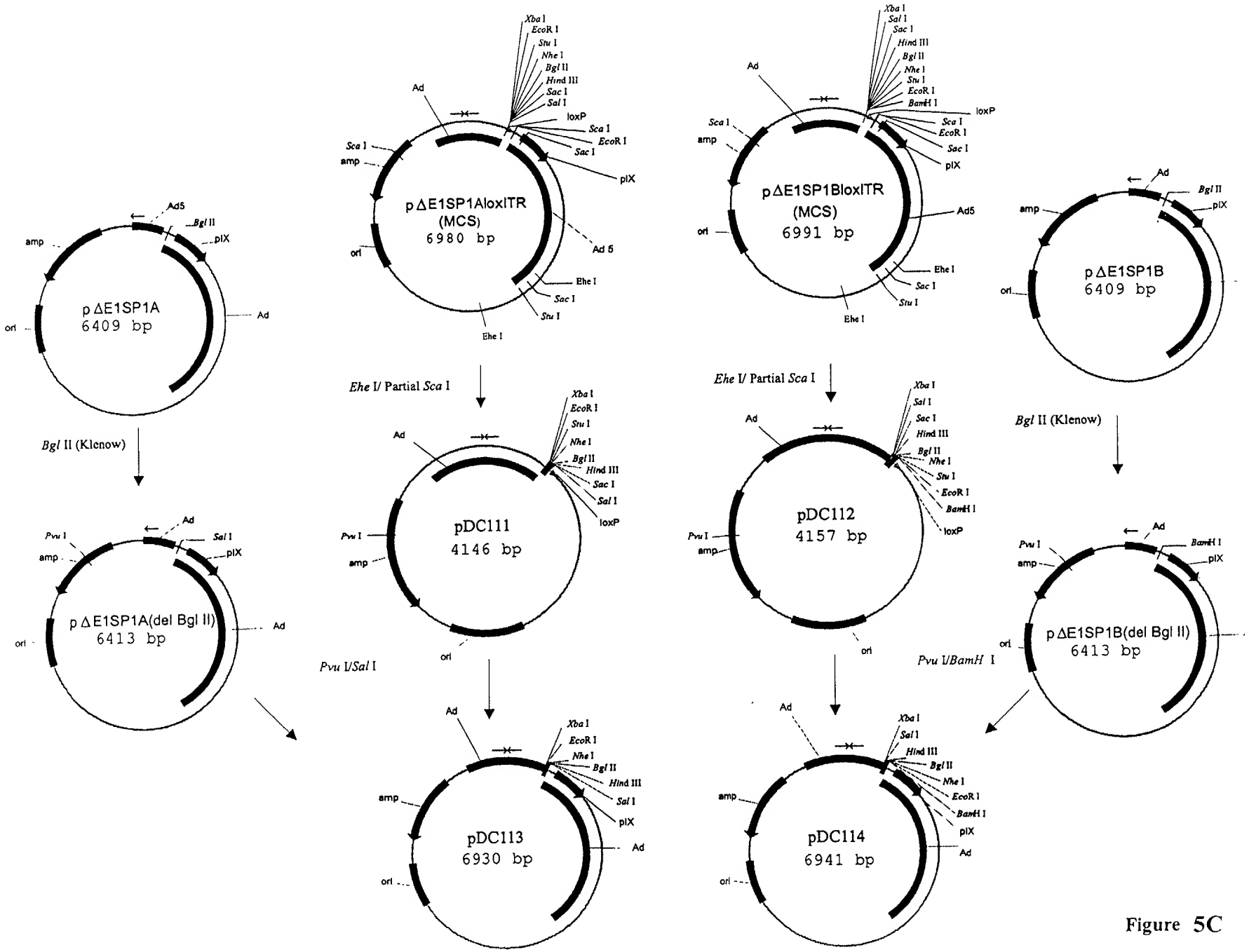
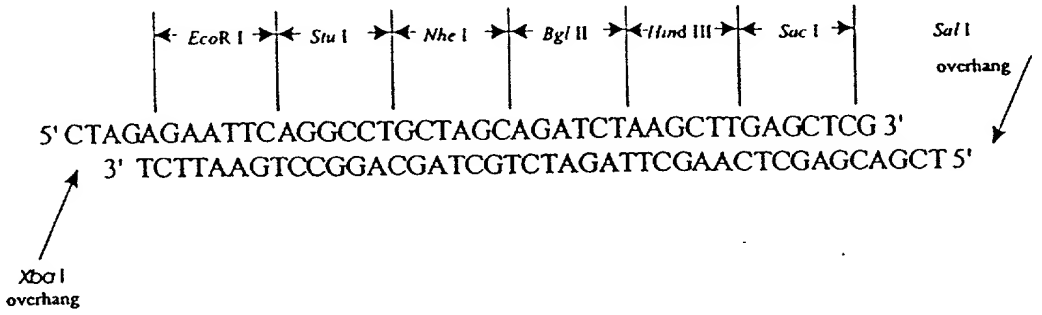


Figure 5C

SEQ. ID. NO.: 12 (AB16853) and SEQ. ID. NO.: 13 (AB16854)



SEQ. ID. NO.: 14 (AB16855) and SEQ. ID. NO.: 15 (AB16856)

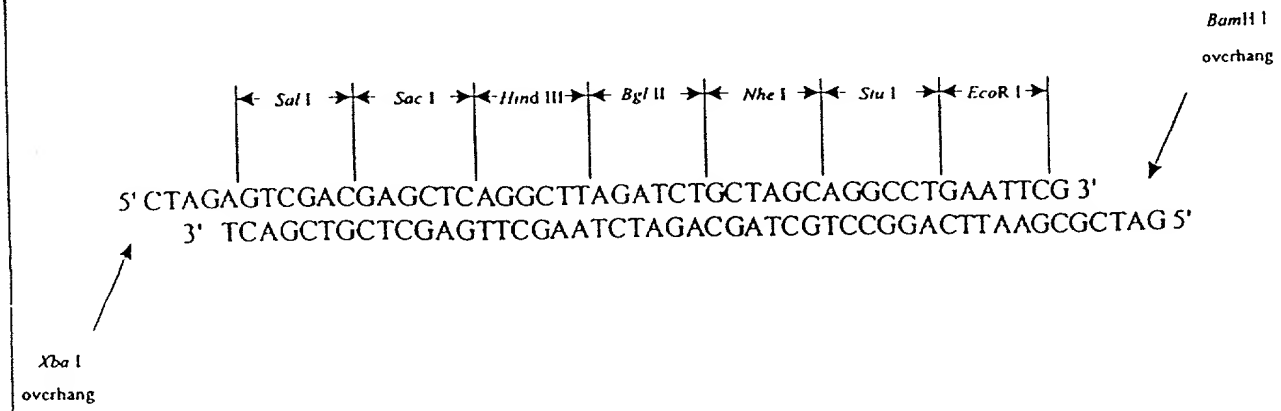


Figure 5D

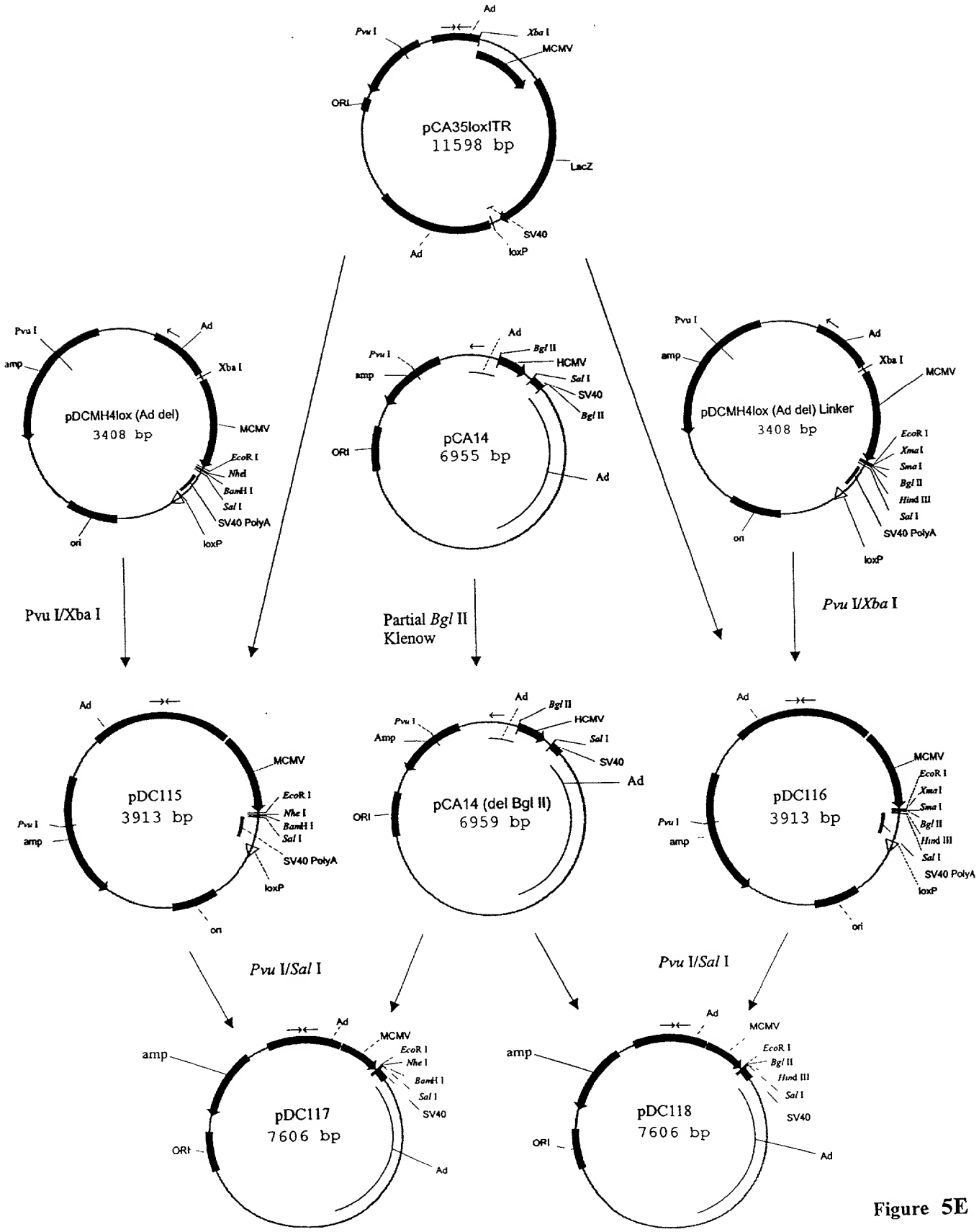

pCA35loxITR
11598 bp
Ad
Pvu I
Xba I
MCMV
ORI
LacZ
SV40
loxP
pDCMH4lox (Ad del)
3408 bp
amp
Xba I
MCMV
EcoR I
Nhe I
BamH I
Sal I
SV40 PolyA
ori
loxP
pCA14
6955 bp
Bgl II
HCMV
Sal I
SV40
pDCMH4lox (Ad del) Linker
3408 bp
EcoR I
Xma I
Sma I
Bgl II
Hind III
Sal I
Pvu I/Xba I
Partial Bgl II
Klenow
Pvu I/Xba I
pDC115
3913 bp
pCA14 (del Bgl II)
6959 bp
pDC116
3913 bp
Pvu I/Sal I
Pvu I/Sal I
pDC117
7606 bp
pDC118
7606 bp

Figure 5E

CONSTRUCTION OF pMH4LOX, pMH4LOXΔ and pMH4LOXΔLINK SHUTTLE PLASMIDS FOR RESCUE OF EXPRESSION CASSETTES

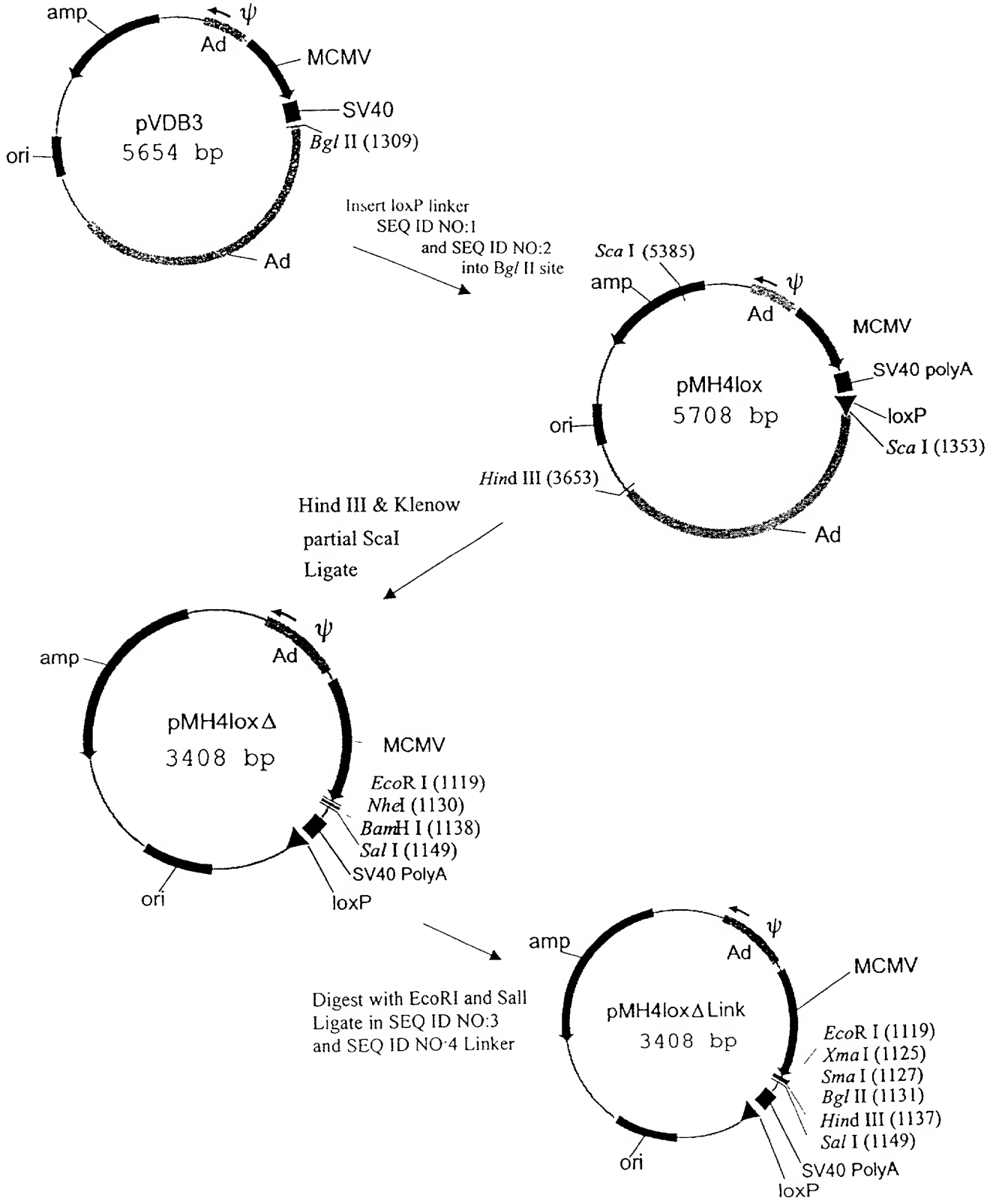


Fig. 6A

CONSTRUCTION OF A SHUTTLE PLASMID CONTAINING A pUC DERIVED ORIGIN

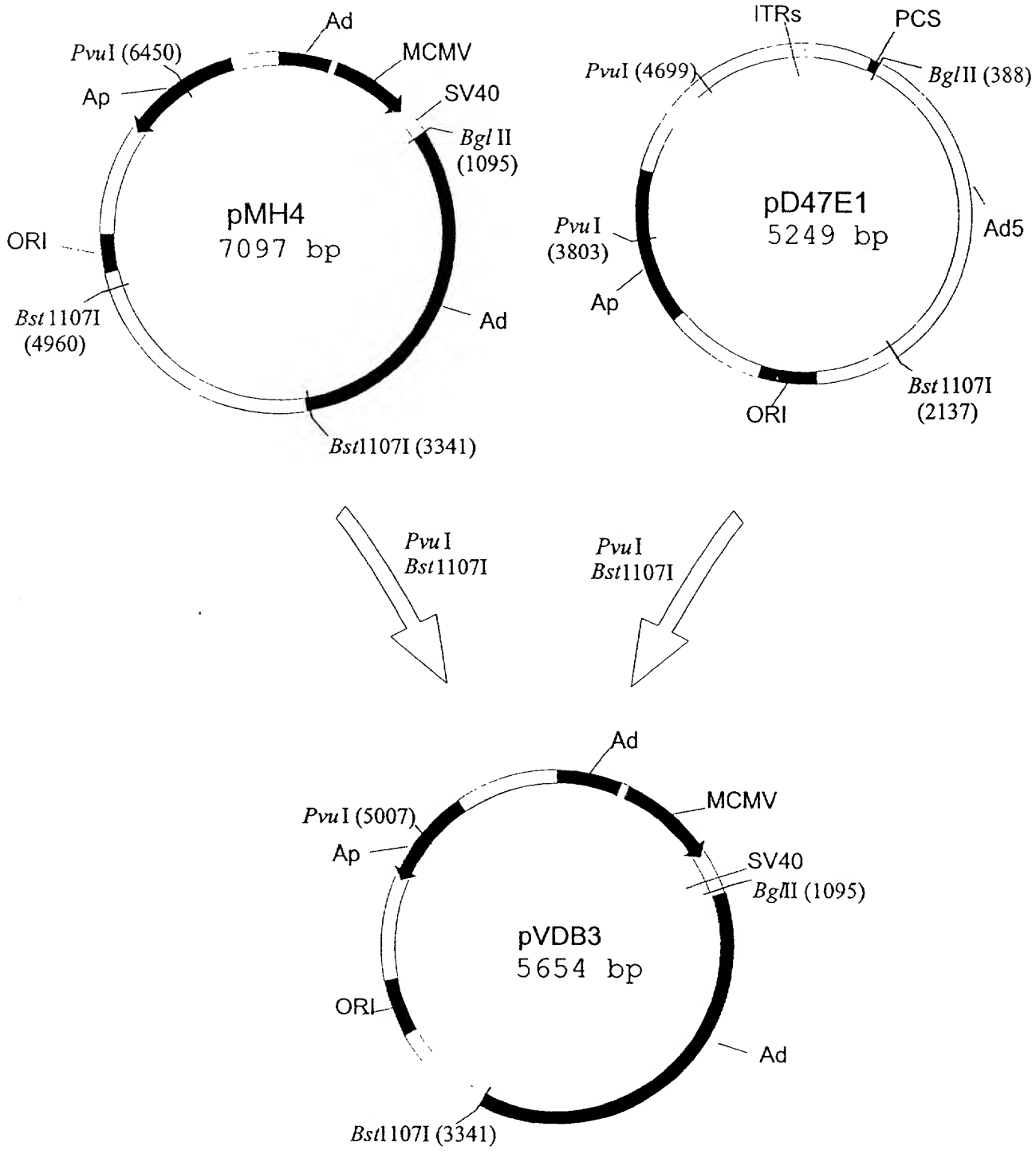


Fig. 6B

CONSTRUCTION OF HCMV loxP PLASMIDS FOR RESCUE OF EXPRESSION CASSETTES

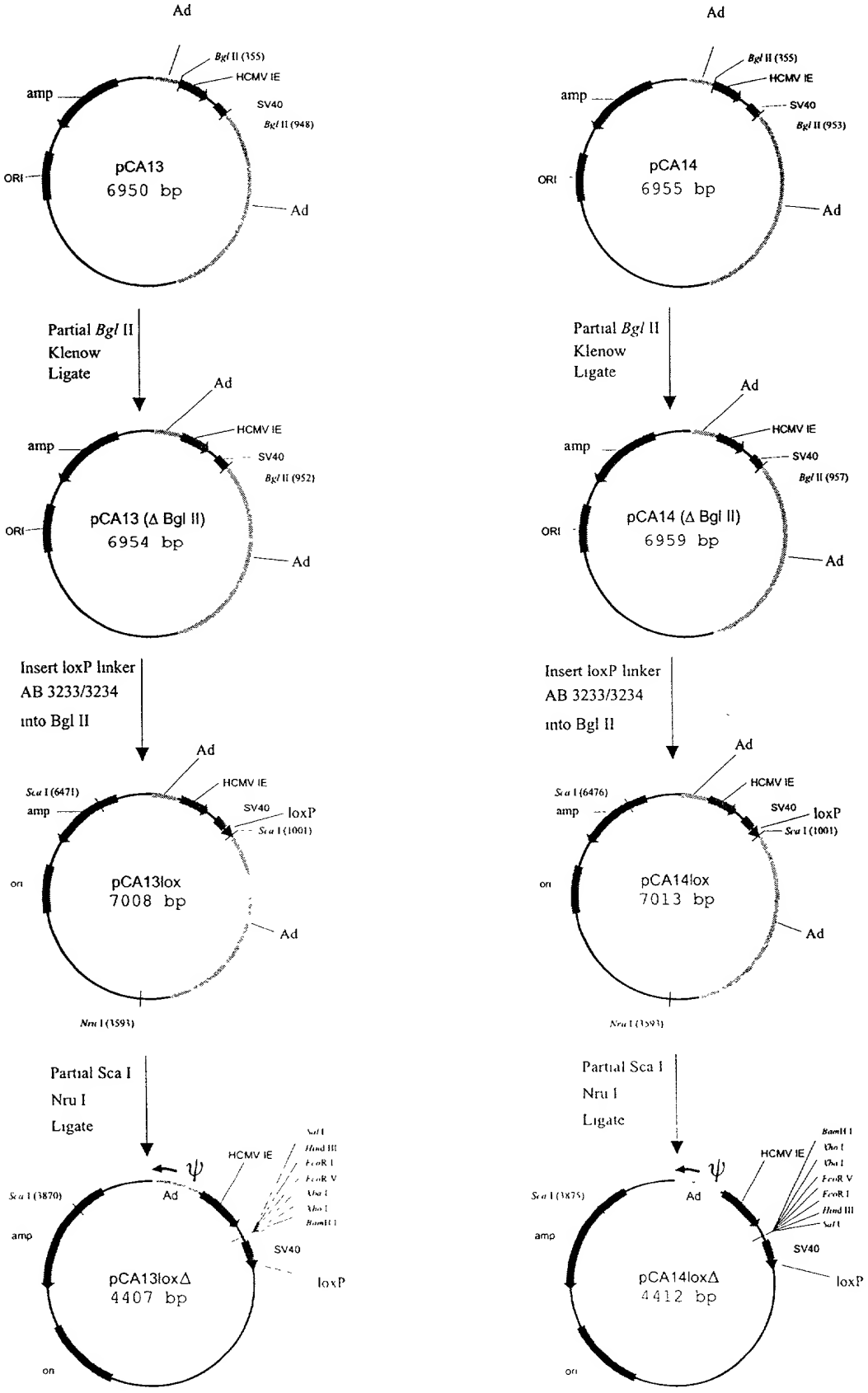


Fig. 7

CONSTRUCTION OF pCA36LOX and pCA36LOXΔ SHUTTLE PLASMIDS FOR RESCUE OF LACZ

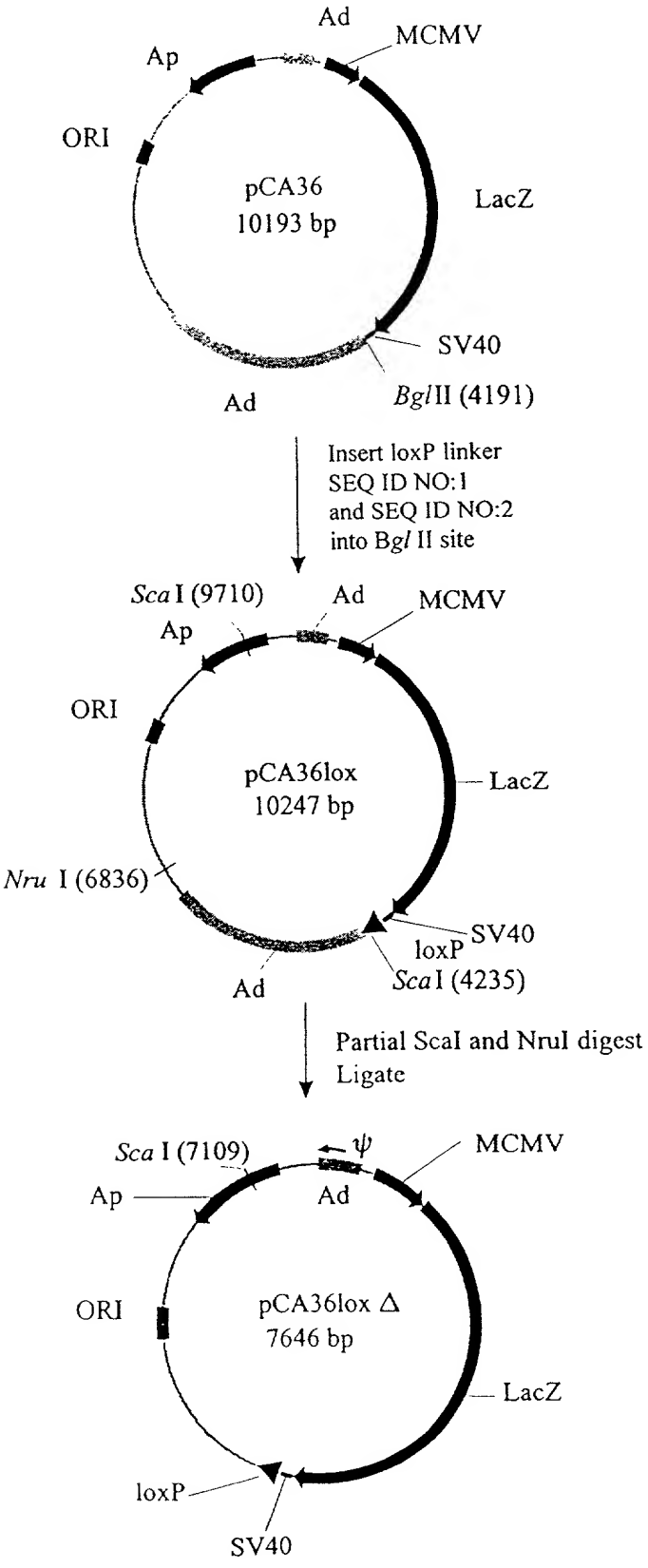
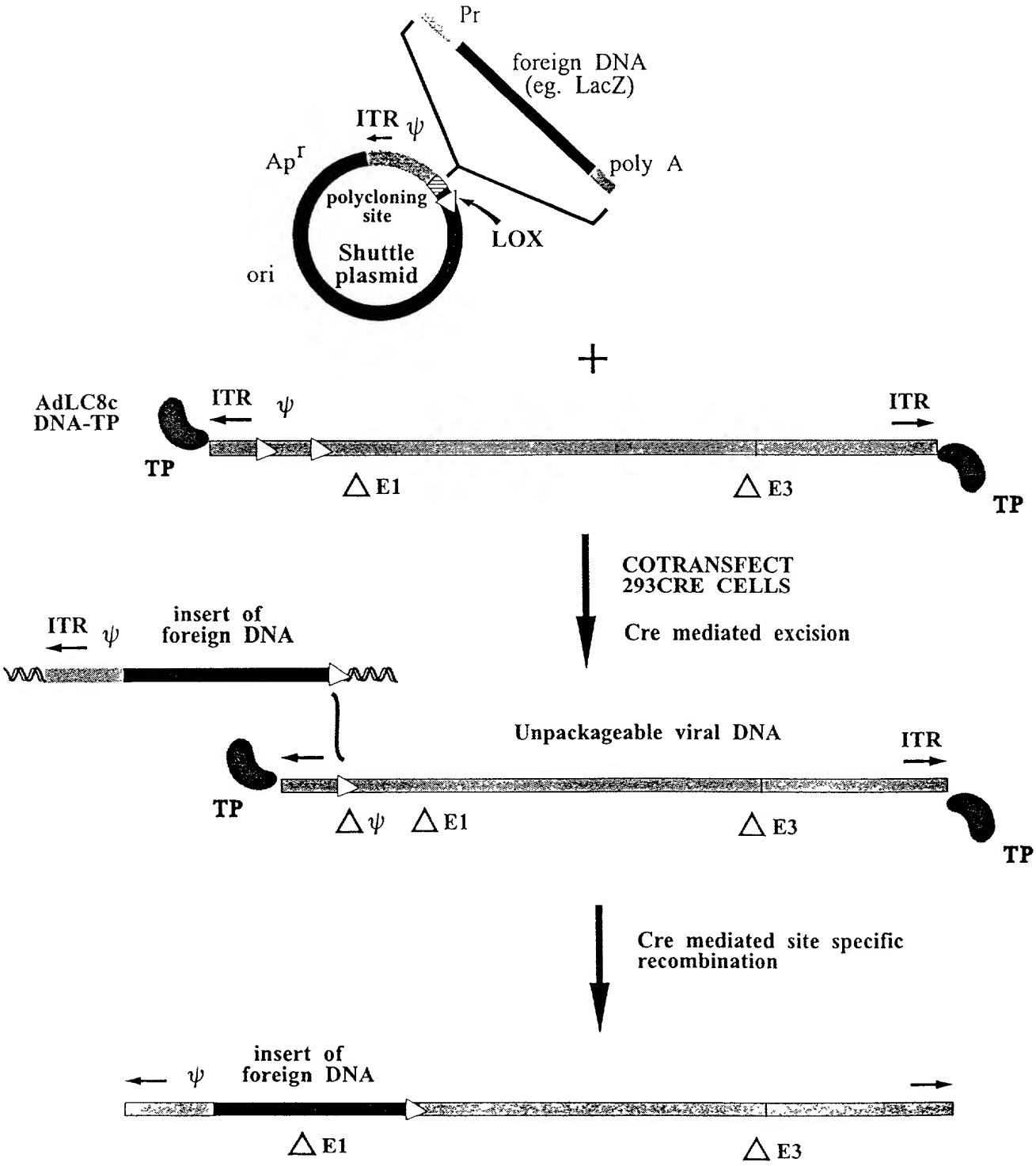


Fig. 8A

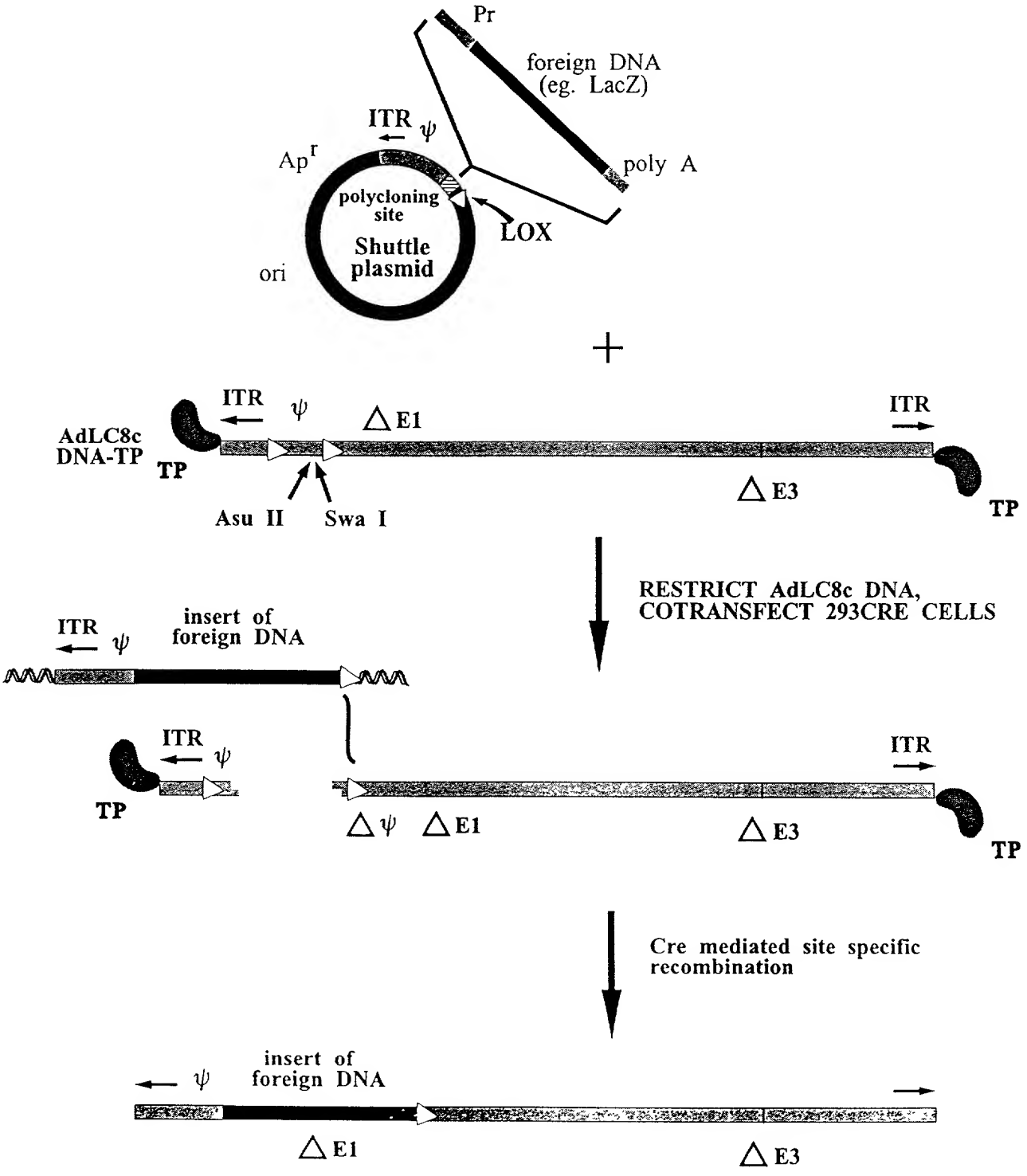
Cotransfection of 293Cre cells with AdLC8c DNA-TP and a shuttle plasmid containing a loxP site for generation of Ad expression vectors



DEFECTIVE VIRAL VECTOR

Fig. 8B

Cotransfection of 293Cre cells with restricted AdLC8c DNA-TP and loxP shuttle plasmid for generation of Ad expression vectors



DEFECTIVE VIRAL VECTOR

Fig. 8C

CONSTRUCTION OF SHUTTLE PLASMIDS EXPRESSING Cre

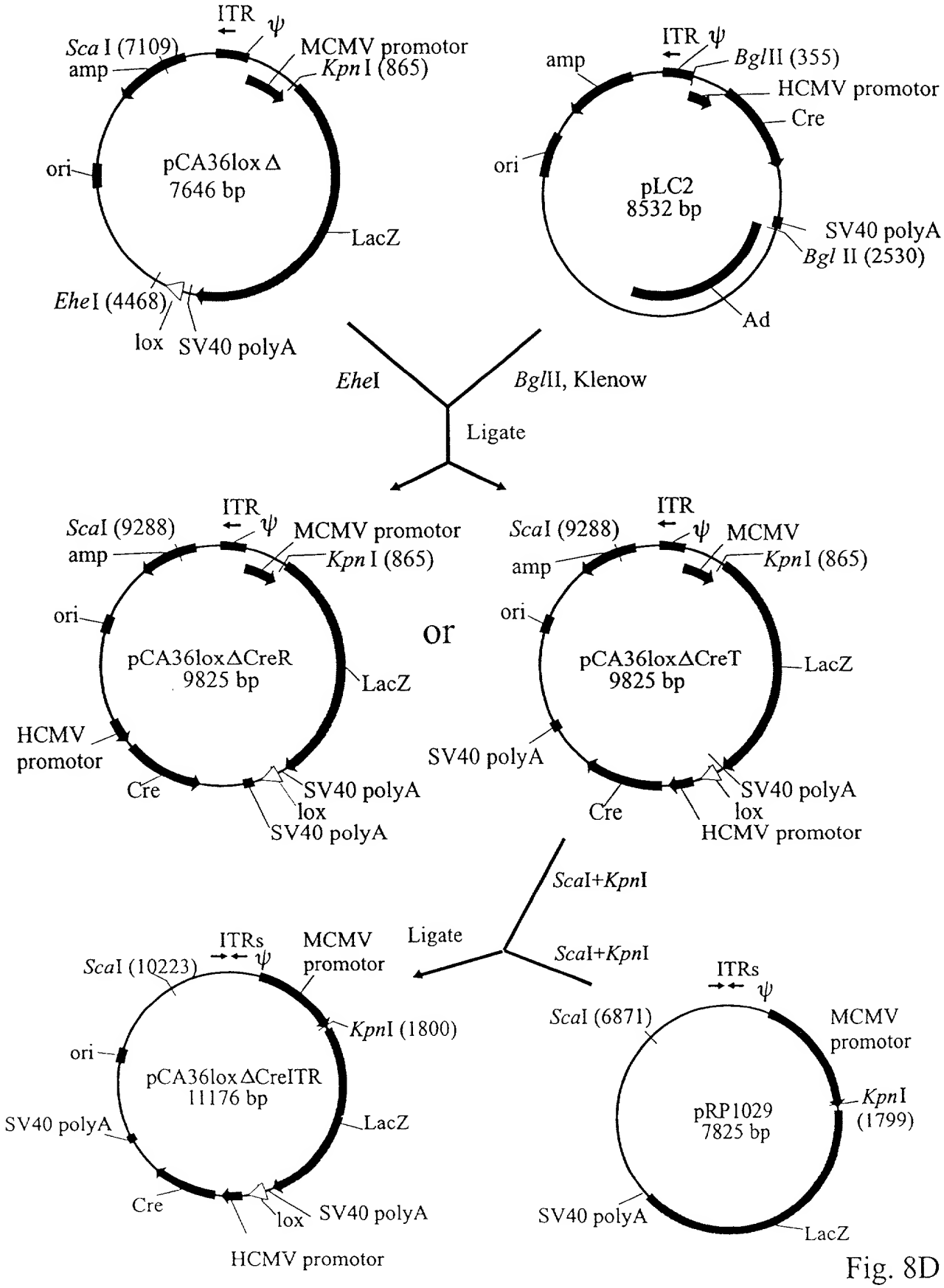
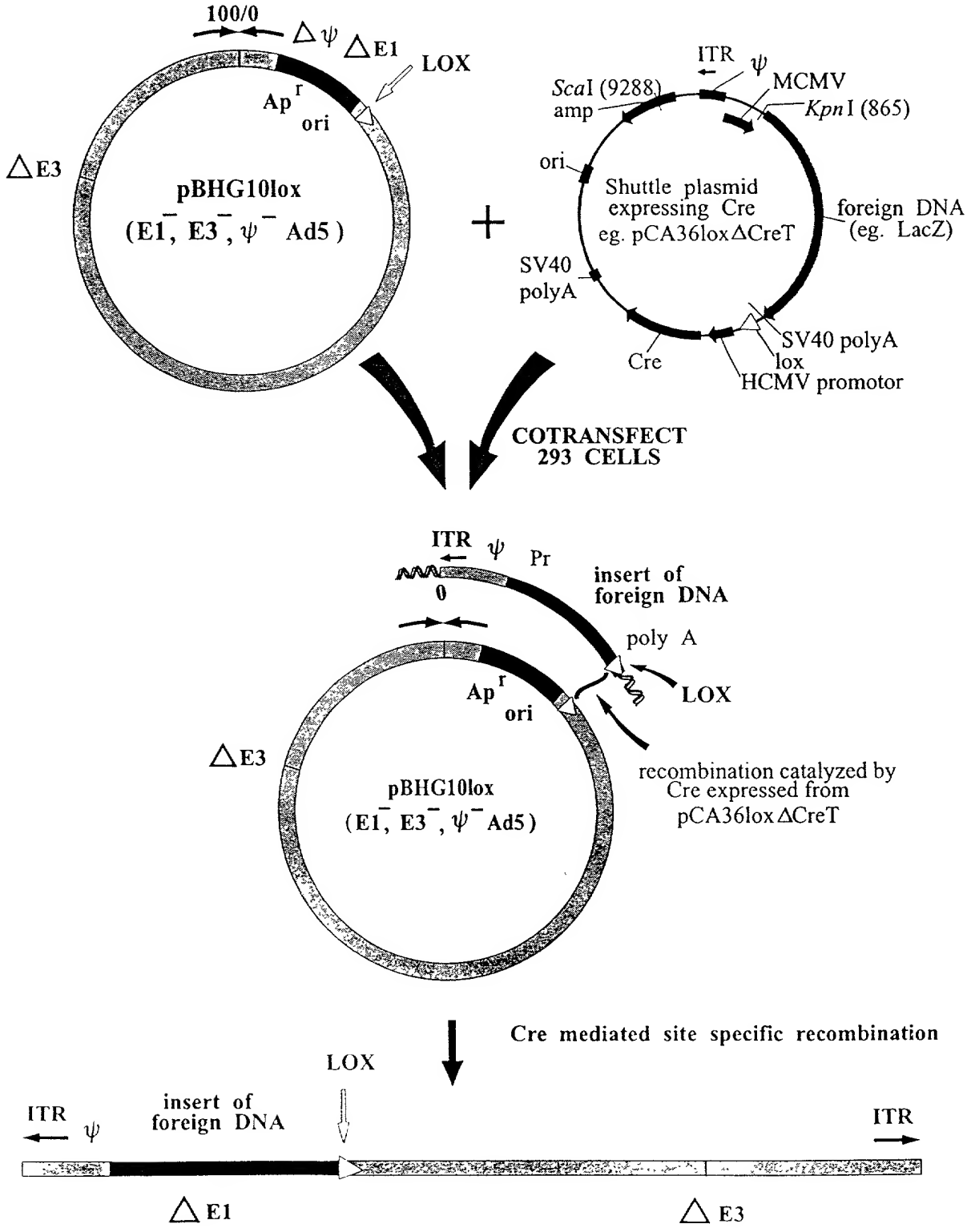


Fig. 8D

Cotransfection of 293 cells with pBHG10lox and a "Lox" shuttle plasmid expressing Cre for generation of Ad expression vectors



DEFECTIVE VIRAL VECTOR

Fig. 8E

CONSTRUCTION OF Ad GENOMIC PLASMID ENCODING CRE

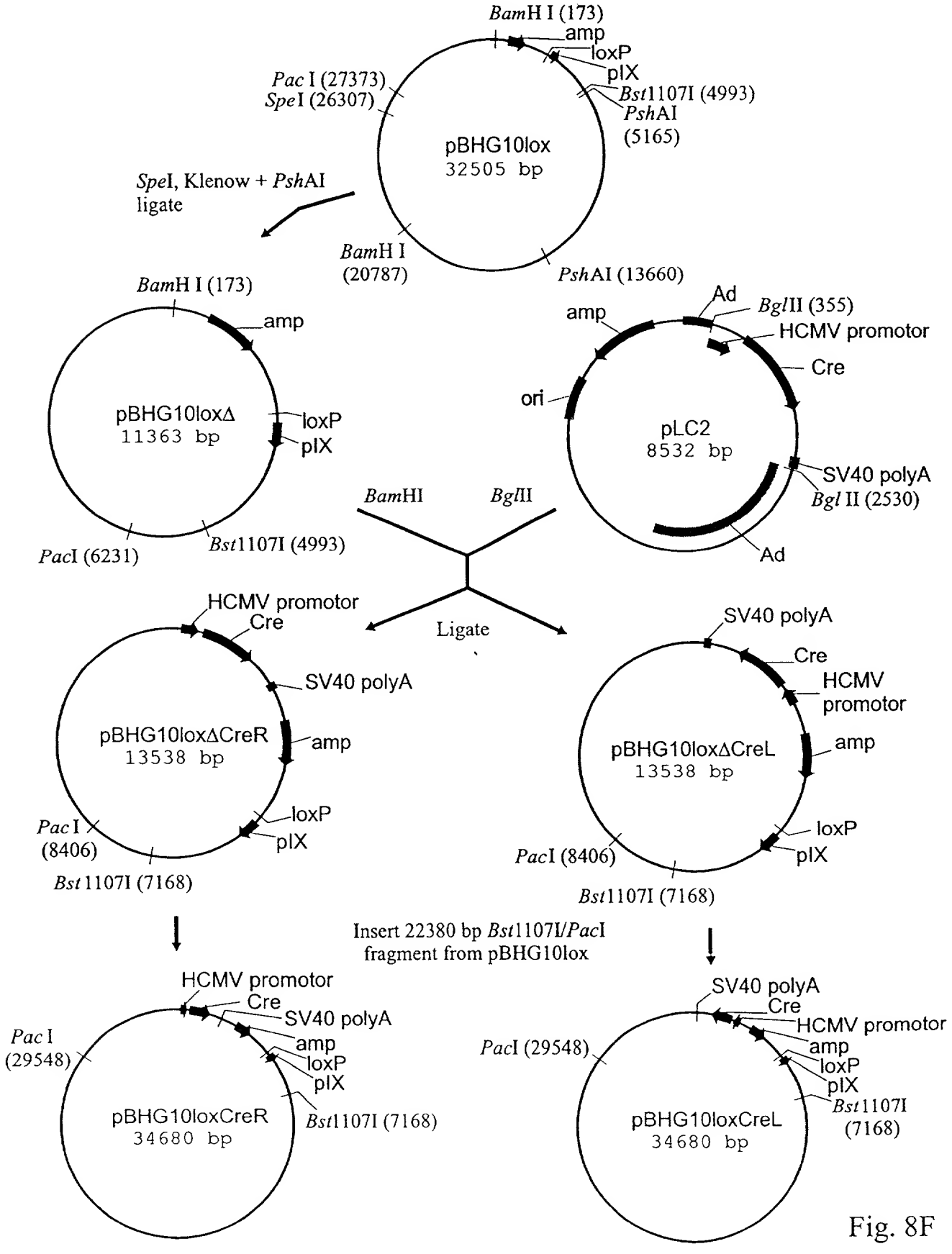
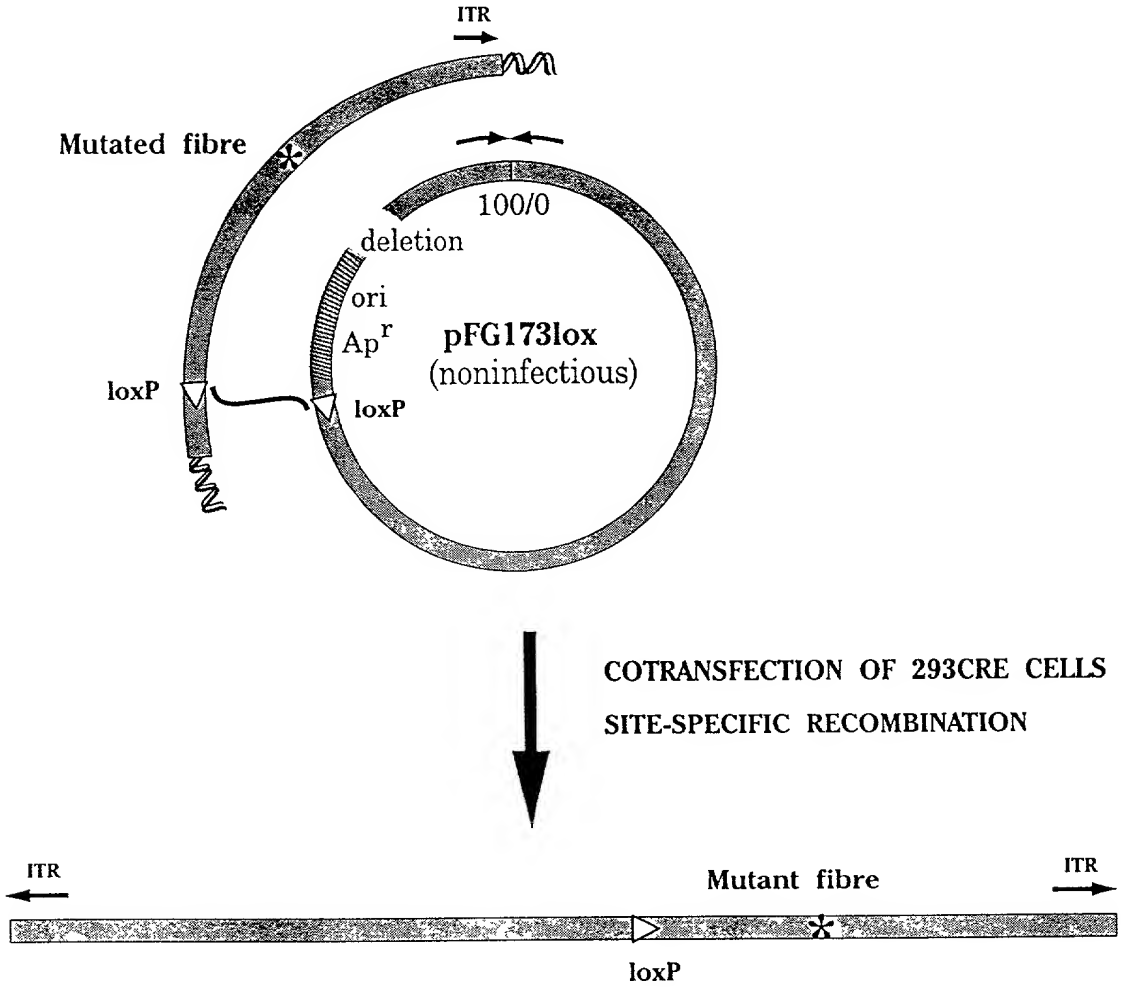


Fig. 8F

RESCUE OF FIBRE MUTATIONS USING CRE/LOX RECOMBINATION



NONDEFECTIVE ($E1^+$) VIRUS WITH MUTATED FIBRE GENE

Fig. 9A

CONSTRUCTION OF pAB14lox △

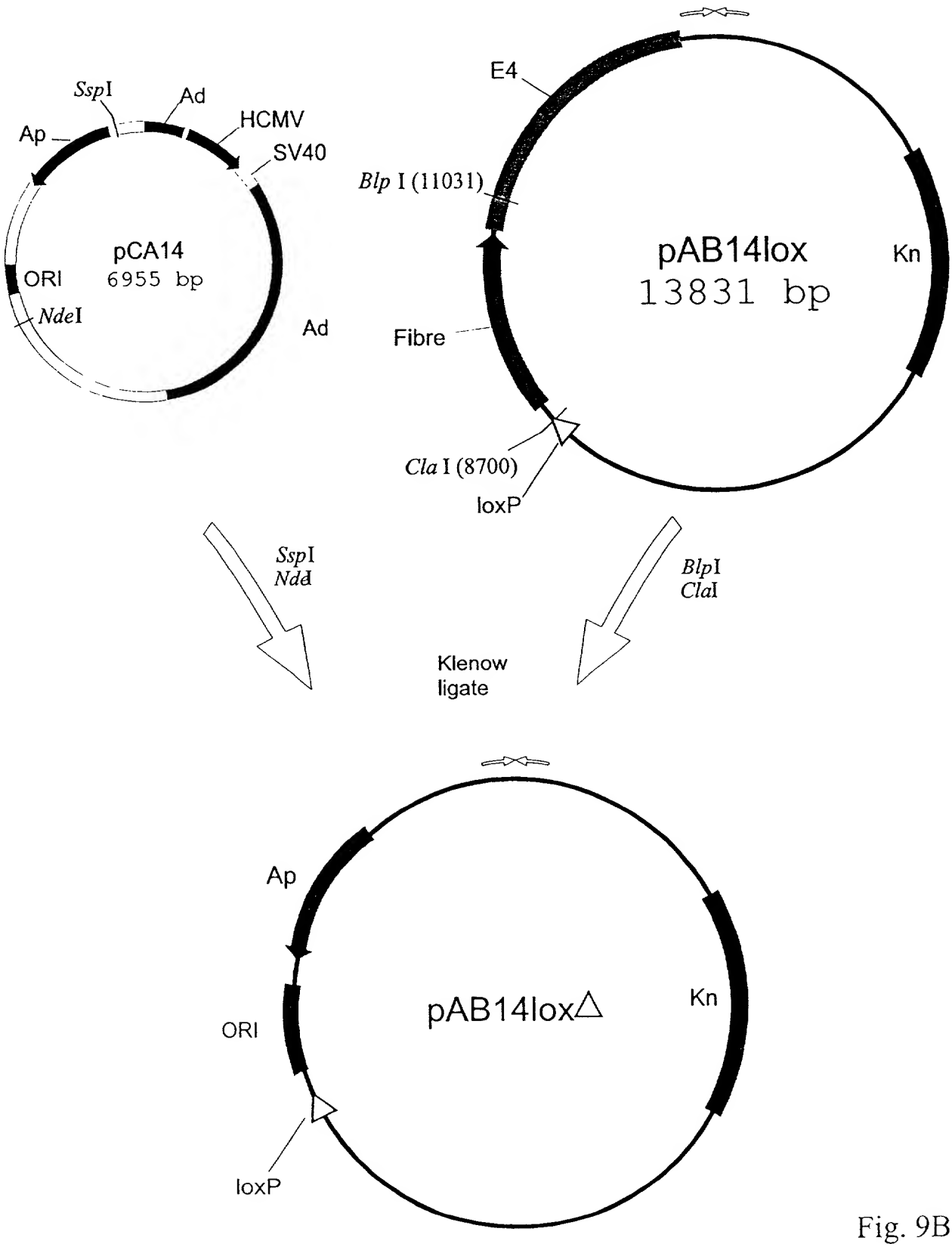
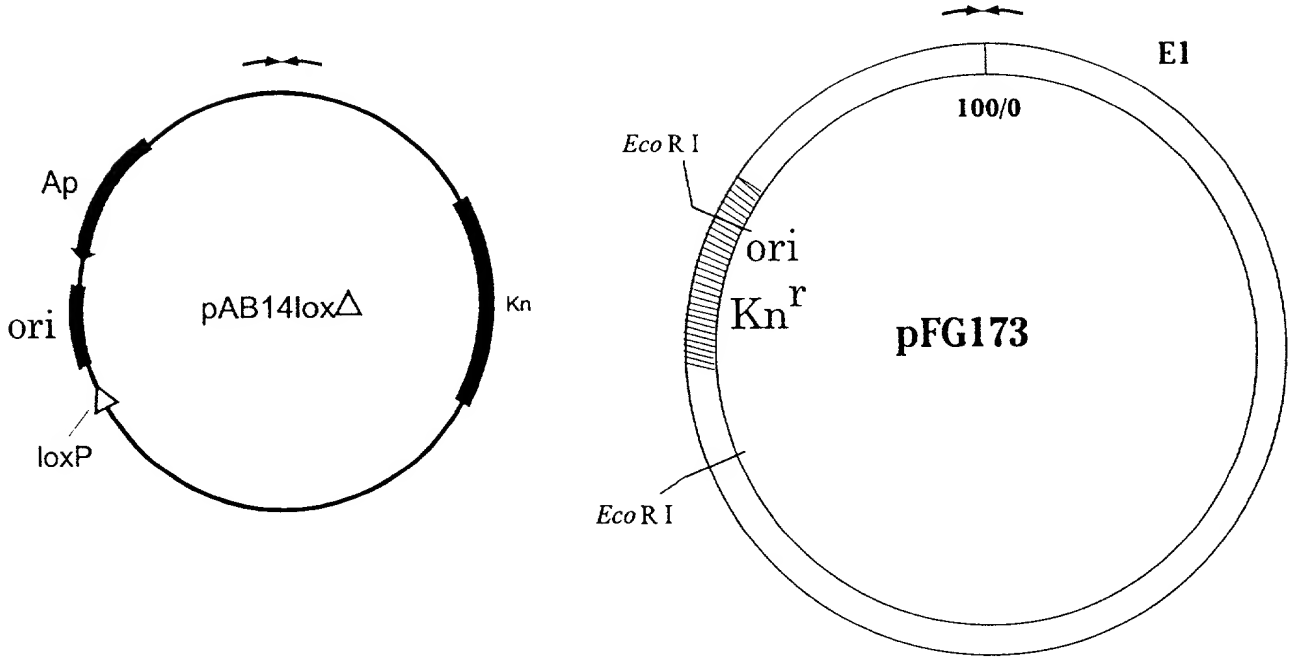


Fig. 9B

CONSTRUCTION OF pFG173lox



Restriction, transformation of E. coli, homologous recombination

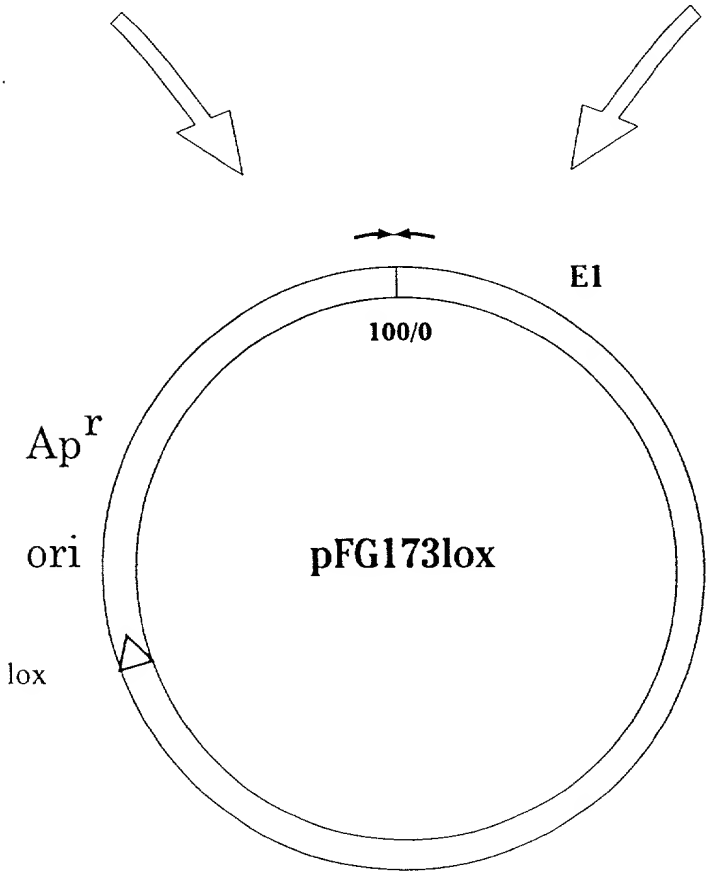


Fig. 9C

CONSTRUCTION OF pFG23dX1lox AND pFG23dX1loxc FOR RESCUE OF MUTANT FIBRE INTO AD VIRUS

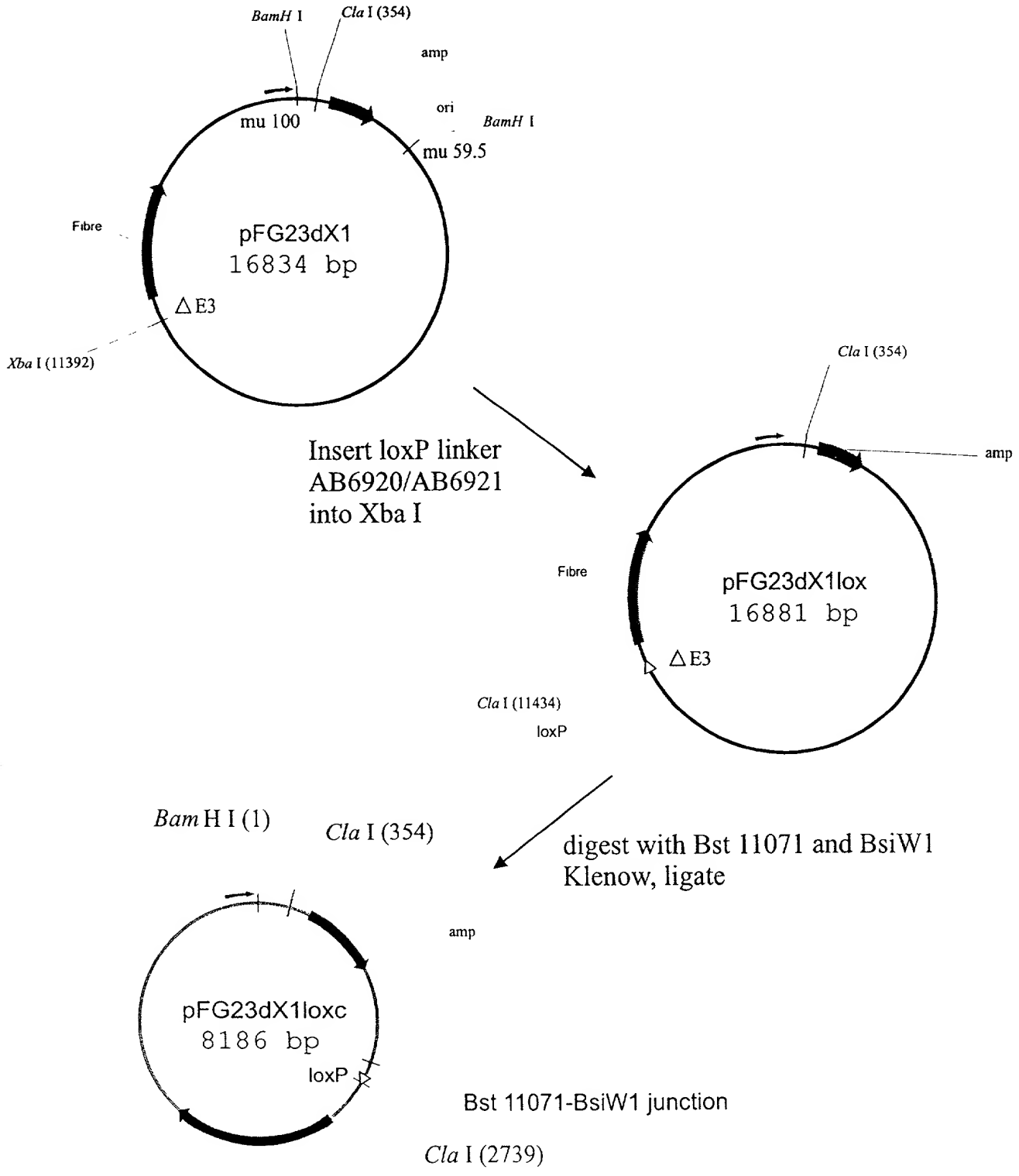


Fig. 10

A PLASMID FOR RESCUE OF A FOREIGN DNA INTO AD VIRUS

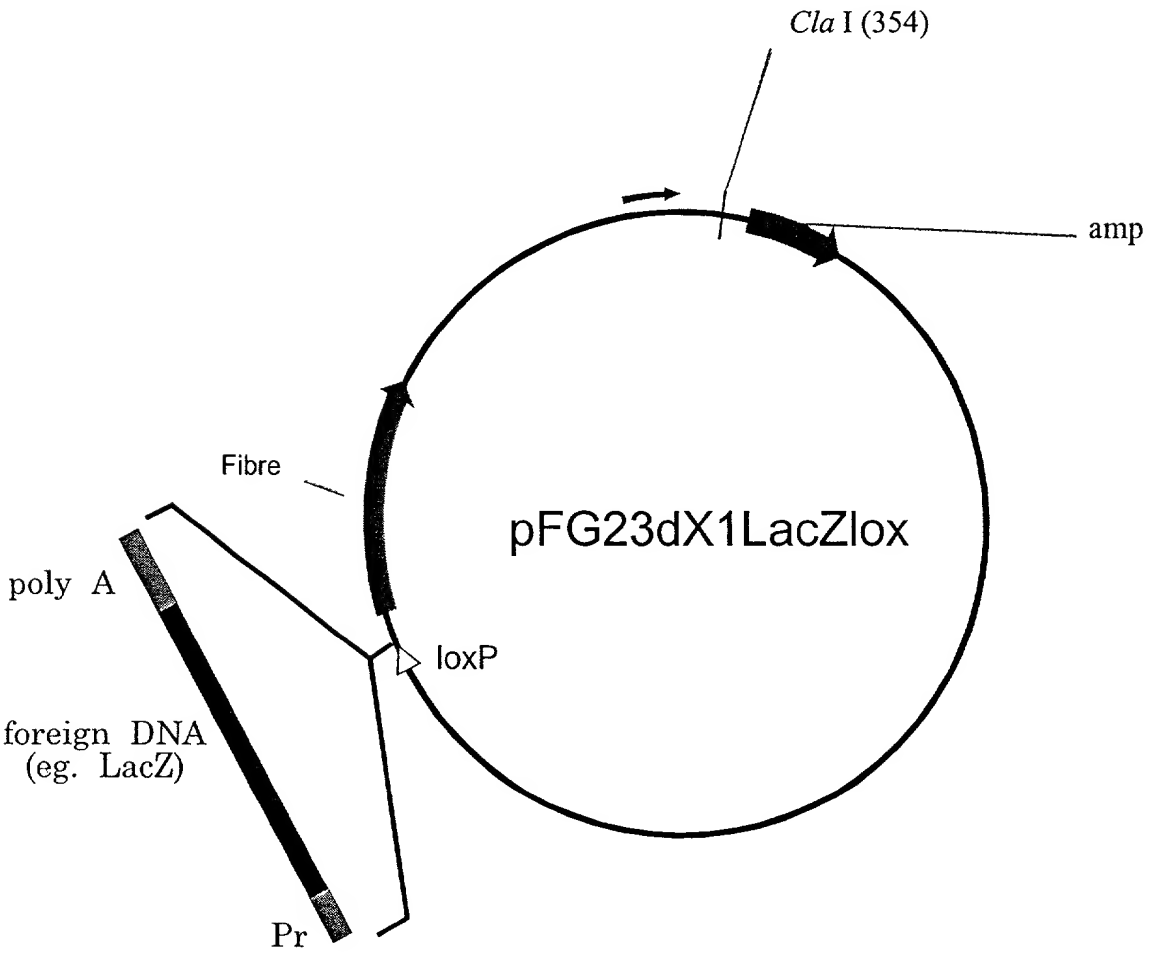
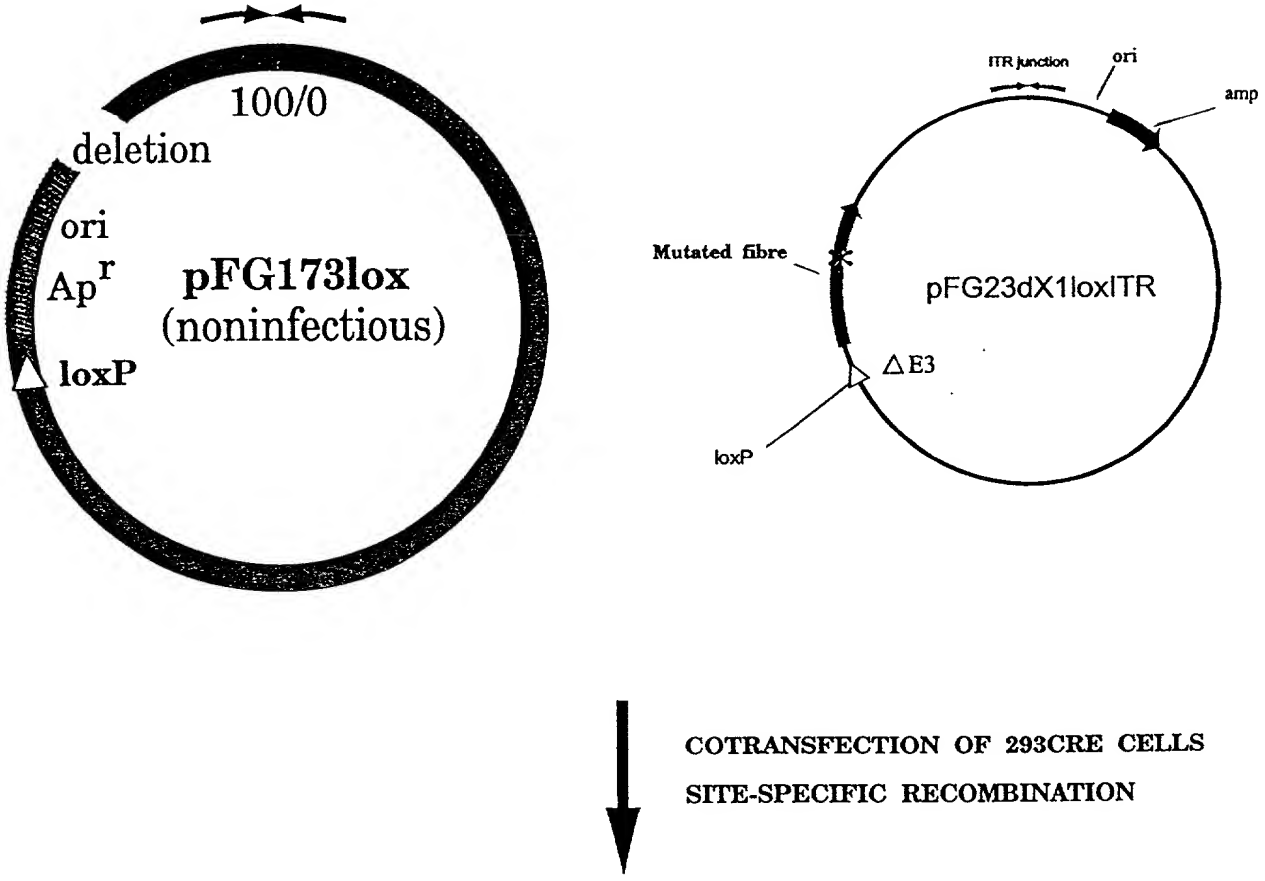


Fig. **11A**

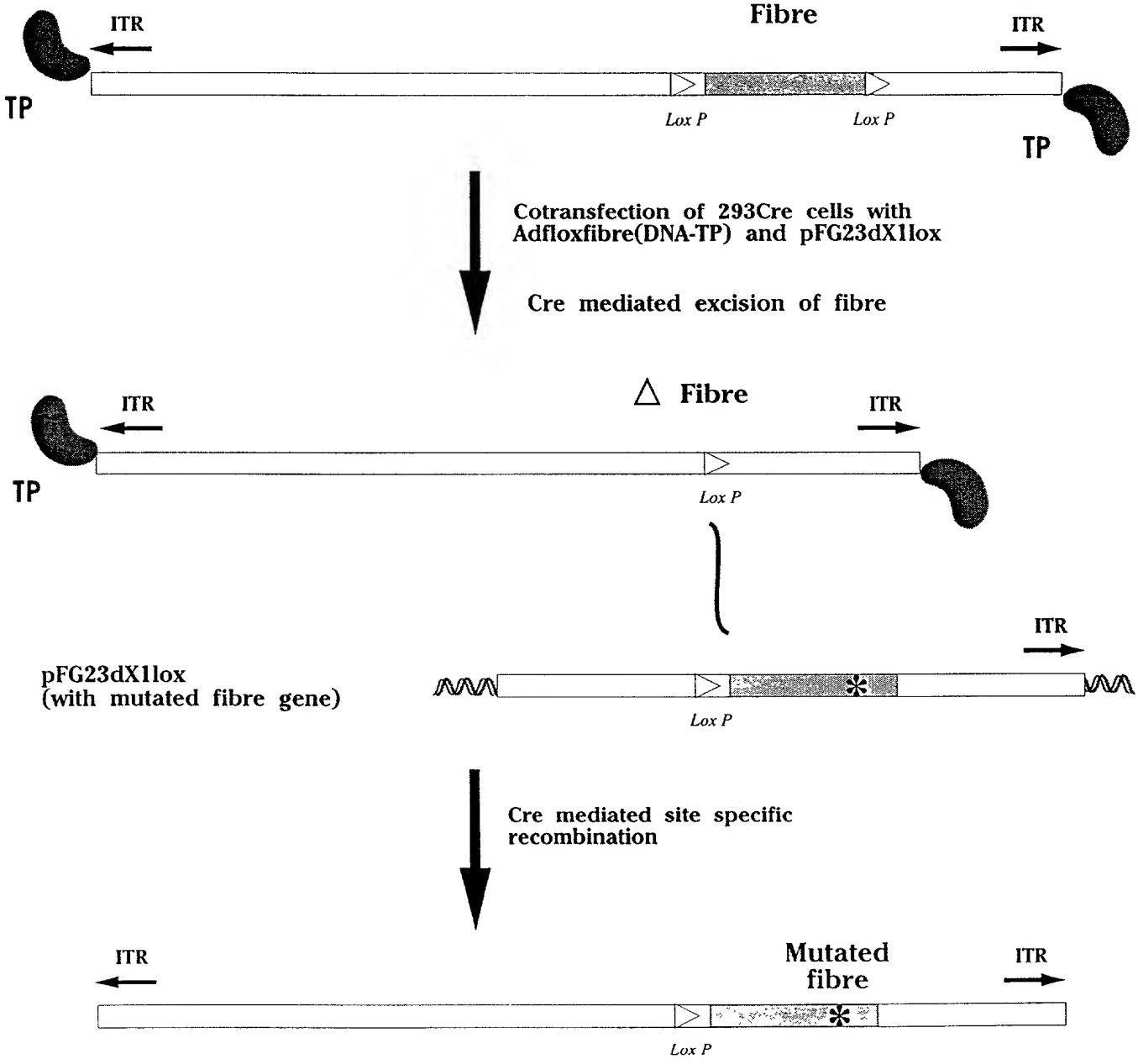
RESCUE OF FIBRE MUTATIONS USING CRE/LOX RECOMBINATION



NONDEFECTIVE ($E1^+$) VIRUS WITH MUTATED FIBRE GENE

FIGURE 11B

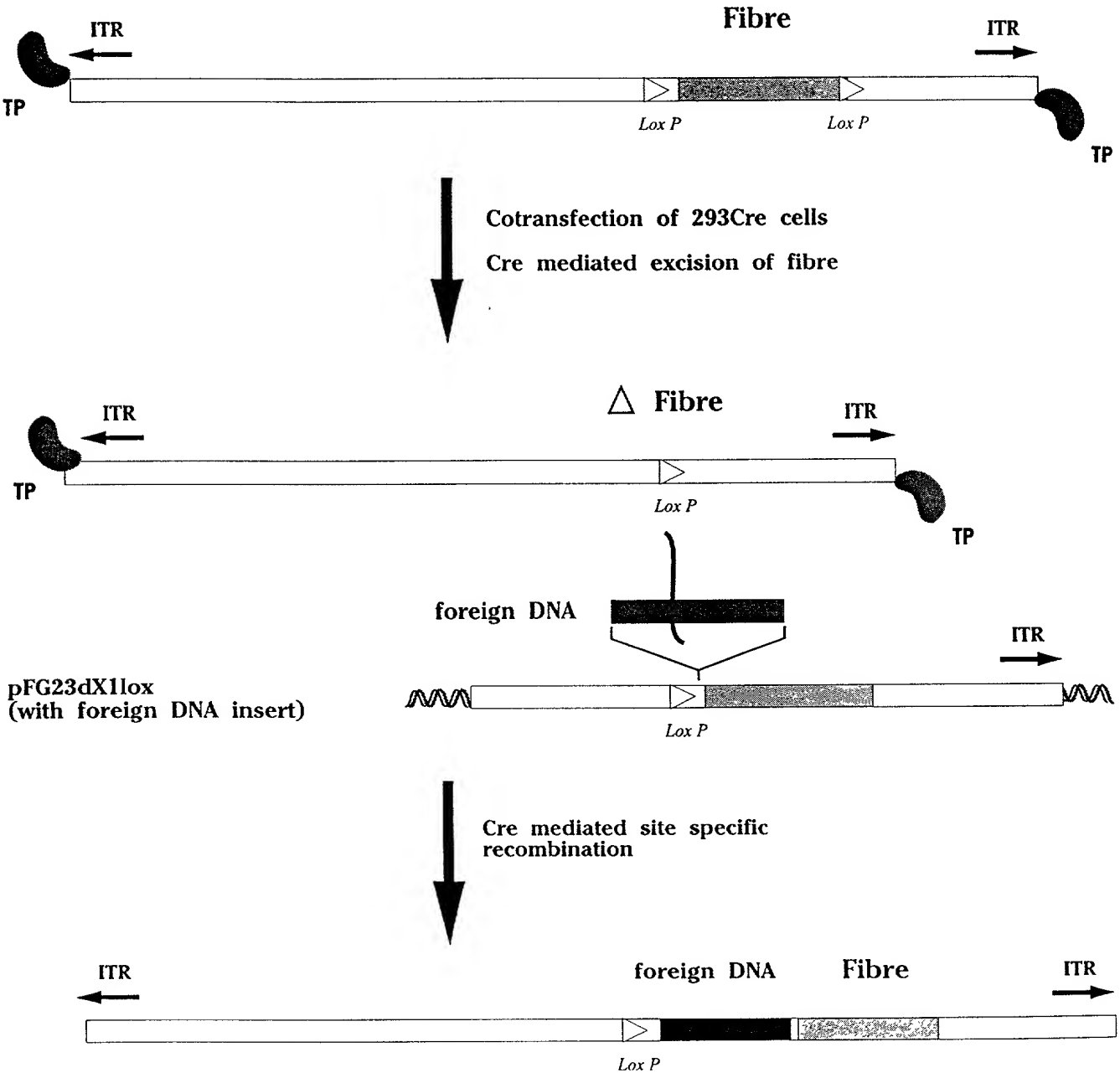
Isolation of a virus containing a mutant fibre gene by Cre-lox recombination using DNA-TP and cotransfection



RECOMBINANT VIRUS CONTAINING A MUTATED FIBRE GENE

Fig. 12

Isolation of a virus containing a foreign DNA insert upstream of the fibre gene by Cre-lox recombination



RECOMBINANT VIRUS CONTAINING AN INSERT OF FOREIGN DNA UPSTREAM OF THE FIBRE GENE

Fig. 13

CONSTRUCTION OF pAB14FL0X FOR ISOLATION OF AN AD VIRUS WITH A FLOXED FIBRE GENE

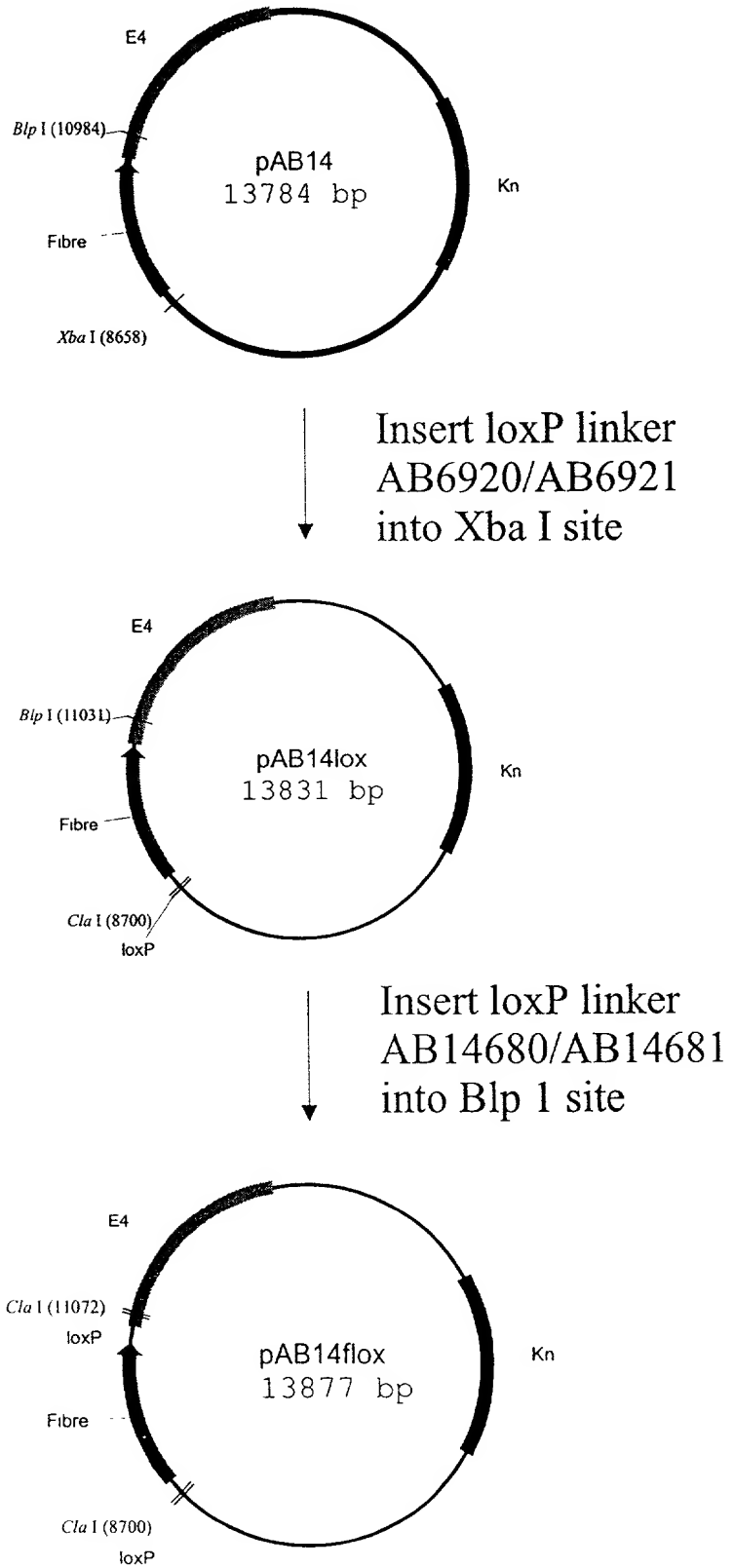
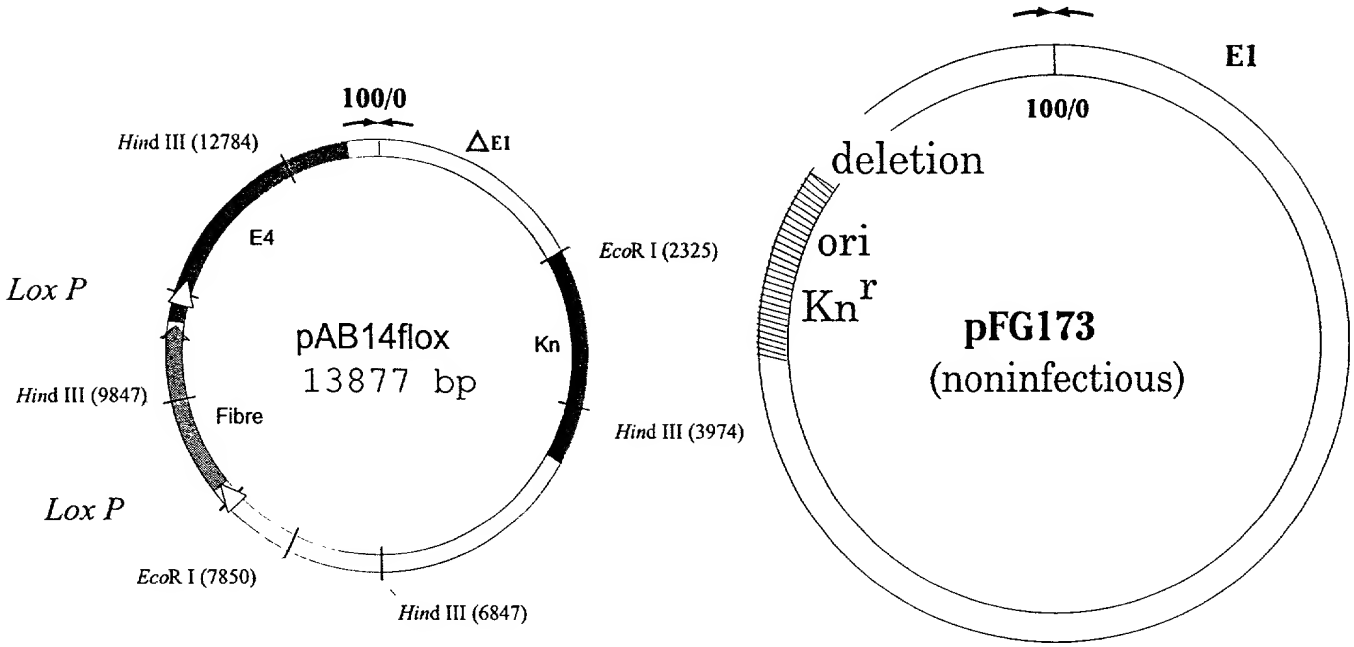


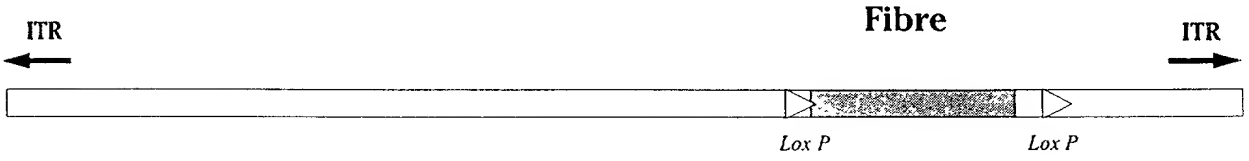
Fig. 14

Isolation of a virus containing a fibre gene with flanking lox P sites.



COTRANSFECTION OF 293 CELLS

HOMOLOGOUS RECOMBINATION



NONDEFECTIVE ($E1^+$) VIRUS (ADFLOXFIBRE) CONTAINING A FLOXED FIBRE GENE

Fig.15